THE
AMERICAN NATURALIST

VOL. LV. *January–February, 1921* NO. 636

GENETIC SEGREGATION[1]

W. BATESON, M.A., F.R.S.

DIRECTOR OF THE JOHN INNES HORTICULTURAL INSTITUTION

THE later developments of Mendelian analysis have been in the main an attempt to elucidate the scope and nature of segregation. Mendel proved the existence of characters determined by integral or unit factors. Their integrity is maintained by segregation, the capacity, namely, to separate unimpaired after combination with their opposites. Our first aim has been to discover specifically what characters behave in this way, whether there is any limit to the scope of segregation, or any characters or classes of characters which are determined by elements unable to segregate simply. The second object has been to decide the time and place in the various life-cycles at which segregation occurs. It is with the latter problem that I propose to deal more particularly in this lecture, but a brief consideration of the range of characters, subject to segregation, is appropriate.

1. THE SCOPE OF SEGREGATION

Of the classes of features by which animals and plants are distinguished, most have now been shown to be dependent on segregable elements. It is perhaps noticeable that we have no quite clear proof that the factors governing differences in number, meristic characters in the strict sense, commonly behave so simply as those determining other characteristics. There are examples of parts repeated in series, such as the extra toe of the

[1] Croonian Lecture delivered before the Royal Society of London, June 17, 1920, and printed in the *Proceedings*.

68555

fowl (a dominant) and the leaf of the monophyllous strawberry with a single leaflet (a recessive) which have a factorial inheritance, but the resulting terms, especially the heterozygotes, are indefinite. In the polydactylous fowl, for example, the heterozygote has a great variety of shapes. The hind toe is most often represented by two sub-equal digits, but the duplicity may be so slight as to appear externally only as a division of the nail. It may also assume another very different form, in which there is only a single many-jointed digit representing the usual pair. In the monophyllous strawberry the homozygous recessive, whether before or after immediate extraction from the heterozygote, shows fluctuations to a 2- or 3-leaved condition.[2] Such fluctuations are common among forms distinguished by meristic peculiarities. There are not the uniformity and simplicity which are such striking features of variations in color and many other substantive characteristics. The evidence as to meristic differences is, however, still scanty, and it is too soon to decide what importance should be attached to this preliminary impression.

With more confidence we recognize that merely quantitative differences seldom, if ever, have a perfectly simple inheritance. There are two obvious interpretations: (1) that the factors do not usually segregate clean; (2) that the number of factors involved is so large that their effects are masked. Adequate discussion of these two possibilities could only be given at considerable length. On the whole, I incline to the former alternative, but the material for a decision scarcely exists as yet. Certain examples should be noticed in which, though the most obvious differentiating feature is quantitative, the underlying physiological distinction is more probably to be referred to a qualitative factor. Height in certain plants is a good illustration. It is ostensibly a quantitative feature, and the talls segregate clean from the dwarfs. But in various cases, *e.g.*, peas and sweet peas, the dwarfs are also a darker green. The dwarf of *Cam-*

[2] Richardson, C. W., *Jour. Gen.*, Vol. 3, p. 171 (1914).

panula persicifolia, especially (about 8 inches high), is a plant so strikingly different from the type (2–3 feet high), that it is sold as a species, "*C. nitida.*" The leaves of the dwarf are an intense, dark green.[3] This variety is, of course, a recessive and segregates without intermediates, yet, if the qualitative distinctions were less evident it might easily be classed as a variety in quantity. But the critical distinction is certainly qualitative and the great difference in size is consequential. Though in such cases segregation is complete, it may almost be said to be characteristic of purely quantitative distinctions that one or other of the original parental types fails to reappear in its extreme form after a cross. So usual is this feature in quantitative segregation that the phenomenon must have special significance.

Another fact is beginning to emerge which must contribute to the shaping of a conception of the physiological nature of segregation. We have learnt that groups or complexes of factors may segregate whole. To such a complex the distinction in sex is due, but in certain cases it may break up. The occurrence of a large spur in fertile hens (Leghorns for instance) must be regarded as due to the absence of that part of the sex-complex which in the normal inhibits the growing of the spur. In ordinary fowls the whole inhibiting group remains on the female side, but the spur-inhibiting element can evidently separate from the rest. On the other hand, when in the cocks of certain breeds (as occasionally in Wyandottes) there is no spur, we may perhaps conjecture that this element has been transferred to the male side.[4]

[3] The ovary projects in a curious way above the sepals so much that, were the plant not a *Campanula,* botanists would describe this ovary as superior.

[4] After much experiment the genetics of spur-development are still very obscure. In the course of a long series begun by crossing spurred Leghorn hens with a spurless Wyandotte cock (giving F_1 normal in both sexes), neither form has reappeared in F_2 with its original development. Very rarely a hen with minute spurs has appeared, and occasionally a cock with the spurs sensibly reduced. Nor from spurred hens $\times$ F_1♂ has anything approaching the original types been raised. Conceivably the detached element is able to join again with the test of the female complex.

The presence of the characteristically masculine comb and wattles in the male Sebright which is otherwise henny, shows that another factor similarly detachable governs their development.

To the breaking up of large compound factors the production of intermediate types, such as occur among the color-varieties of plants, is in all likelihood due. The sweet pea and the snapdragon have now an innumerable series of such color-forms which may be represented as having arisen by the disintegration of the various anthocyanins. That, at least, is the simplest way in which their origin can be conceived.

To the final result many qualifying elements contribute, and these may naturally be separate entities. But change in the amount of the same coloring material, and diminution in the mere extent of its distribution, are common features of these graduated series. As the cultural development of the species progresses, more and more of these quantitative intermediates appear, and are selected until a practically continuous series is produced. Although the interrelations of the whole series can be represented by a factorial scheme, the assumption that each of these factors had an aboriginal individuality appears gratuitous. In *Antirrhinum,* for instance, the ordinary self-colored flower segregates as a single unit from the white. But there are "Delila" forms having the "face" colored and the "throat" white. Another variety has the "lip" colored and the peripheral parts white and to this again there is an almost exact counterpart in which the peripheral areas are colored and the lip nearly white, and between these again there are further intergrades. Apart from factors modifying its quality, the color of the whole corolla, segregating as a single entity from the white, would without hesitation be represented as depending on a single factor. Subsequent experience that this entity can break up into an indefinite number of fractions is not evidence that the original representation was wrong. This reasoning applies to a great range of phenomena.

In view of the chromosome theory of linkage, it is therefore worth remarking that we do not find linkage especially frequent among these fractional factors. Have they, then, been distributed among different chromosomes? If in *Antirrhinum,* the color of the face and of the throat were lately parts of a single factor for the total flower-color, would not linkage between them be expected? Nevertheless, in cases of this sort, so far as I know, linkages have not been found with any special frequency.

The segregation of a group of differences—and presumably factors—in combination has lately been shown by Renner[5] to occur with extraordinary frequency in the Œnotheras, and this peculiarity is without doubt at the bottom of the difficulties which have beset the genetic analysis of these plants. The complexes are in several forms or species not borne equally by the two sexes of the same plant, and most of them are unable to exist in the homozygous state. These discoveries greatly elucidate the Œnothera problem. For instance, not only *Œn. lamarckiana,* but *biennis, muricata,* and others also, are not homozygous types, but heterozygotes of a special kind. Consequently, the production by them of "mutants" is not capable of the simple interpretation originally applied to them by de Vries. Renner suggests that the mutants arise owing to some interchange between the complexes which at present we can not investigate, but whatever be the exact manner of their origin we can not regard them as genuine examples of the production of novel forms by a homozygous type.

Before leaving this part of the subject, we may notice that the supposition that segregation is concerned solely with characters of a superficial or trivial nature has been long ago disproved. Baur's Antirrhinums, the study of which was continued by Lotsy, were an excellent demonstration to the contrary, for they provided many illustrations of segregation in features, the "specific value" of which no systematist would question. If further evi-

[5] *Zts. f. ind. Abstammungs- und Vererbungslehre,* XVIII, 1917, p. 121.

dence were needed, it may be found in the fine series of experiments lately published by Heribert-Nilsson[6] in *Salix,* which, contrary to the belief long ago entertained by Wichura, show that, when F_2 families are raised in adequate numbers, very striking segregation occurs in the species-crosses. Many geneticists are inclined to the view that segregable characters should be pictured as implanted on an irreducible base which is outside the scope of segregation, but no means have yet been devised for testing the reality of this conception.

2. *The Moment of Segregation*

The next question is to determine when in the various life-cycles segregation can occur. Obviously it is a phenomenon of cell-division. If we knew nothing of the genetics of plants we might confidently adopt the view which Morgan has so successfully developed, that normal segregation and redistribution happen exclusively in the process of reduction. Though unconvinced, I can not deny that linkage and crossing-over may well be represented provisionally, as effected during synapsis. The scheme previously offered by Punnett and myself as a diagrammatic plan capable of representing these phenomena is certainly far less attractive. There is evidence that in certain plants, *e.g.,* Matthiola, the composition of the families derived from single pods shows very great and perhaps irregular fluctuations, and the normal ratios for those families is only found by taking the average of many, but no sufficient statistical examination of such cases has yet been made. In some suitable case estimations of the offspring derived from individual anthers might be of value in this connection. Renner, by examining the starches of the pollen grains in Œnotheras, has lately made visible that dimorphism, of which we had previously genetic proof, and perhaps this novel and striking observation might be used for the purpose of mapping the distribution of such a character

[6] Lund's "Universitets Arsskrift, N.F.," Avd. 2, Vol. 14, November 28, 1918.

among the pollen grains. Meanwhile, it must be granted that no indication that gametic linkage results from somatic differentiation has yet been obtained.

When, however, we examine the view that linkage of factors is a consequence of their association in a chromosome, we must observe that there is no body of evidence that the number of linkage-systems agrees with that of the chromosomes, a primary postulate of Morgan's theory. *Drosophila* is the only example which has been adequately investigated. The cytological appearances are not readily consistent with the other postulate of Morgan's case, that crossing-over is effected by anastomosis of chromosomes and exchange of materials between them. In our present ignorance of the physical nature of the factors we are not obliged to assume that an actual transference of material is an essential condition for an exchange of properties; but since Morgan's suggestion is made in that form we are bound to notice how difficult it is to interpret the visible phenomena of cytology in accordance with that hypothesis. Without personal familiarity with cytology no one can have a confident opinion. I observe, however, that in his most recent publication on these subjects, E. B. Wilson[7] gives a very emphatic "counsel of caution," remarking that writers on genetics have taken too much for granted, and that for the present the genetic development of the chromosome theory has far outrun the cytological. To a layman the visible appearance of chromosomes is scarcely suggestive of the prodigious material heterogeneity demanded, and the general course of cytological evidence seems to indicate that the rôle of the chromosomes is passive rather than active. Though showing features of regularity, they are capable of very wide variations without transgressing the limits of viability, which could scarcely be the case were every detail in their organization critical. The appearance of chromosomes is not to me suggestive of strings of beads of extreme heterogeneity, but rather of strands of some more

[7] AMER. NAT., p. 210, May–June, 1920.

or less homogeneous substance; and in so far as numerical and geometrical order is exhibited by them, it would, in my opinion, be more proper to compare this regularity with that seen, for example, in drying mud or in the formation of prisms of basalt, than to attribute to it a more fundamental meaning.

Leaving these speculative considerations, and limiting our inquiry to the concrete question, at what moment in the cycle does genetic segregation occur, we reach a perfectly definite answer: that whatever future research may decide as to the occurrence of segregation in animals—which, for aught we know, may always be effected at the reduction division—there is no such limitation in plants. We are now thoroughly familiar with a large group of examples in which the genetic properties of the male and female cells of the same plant are quite different. In these, at all events, the reduction-division can not be the moment of the segregation by which these characters are distributed.

The first case detected was in *Matthiola,* where Miss Saunders' results proved that in the double-throwing singles the pollen carries exclusively doubleness, the eggs being mixed, some single and some double. A similar condition was shown to exist in regard to the cream and white plastids, respectively, the pollen grains bearing exclusively cream. De Vries observed a comparable arrangement among the Œnotheras, and Renner has lately shown that the phenomenon is widely spread in that group, thereby providing a consistent interpretation of much that was formerly obscure in the genetic behavior of these plants. In *Campanula carpatica* Miss Pellew proved that the pollen grains of the hermaphrodite form called *pelviformis* carry exclusively femaleness, and preponderantly white flower-color (the plant being heterozygous for blue). The case of Petunia investigated by Miss Saunders[8] is somewhat peculiar in the fact that in the heterozygous singles the male side carries the dominant singleness only, since in those in-

[8] *Jour. Gen.,* Vol. 1, p. 57 (1910).

stances to which the conception of dominance can be applied, it is the male which commonly carries the recessive. Segregation of these characters can not in plants so organized be supposed to take place later than the constitution of the male and female organs, and therefore the reduction division can not be the one critical moment. The suggestion has been made that germ-cells of the missing kinds may be in reality formed and perish before reaching a functional stage. As regards the Œnotheras, where shriveled pollen grains abound, this conjecture is very plausible and probably correct; but when, as in the other cases here quoted, the pollen grains are uniformly sound, the hypothesis is inapplicable and without evidential support. Moreover, even if it were true that certain classes of germ-cells perish in one or other of the sexes, that would hardly alleviate the difficulty, for this differential viability would remain to be accounted for, being itself a phenomenon of segregation.

Begonia Davisii[9] is another curious illustration in which segregation must occur even earlier. This plant is a wild, true-breeding species, with ordinary single flowers. All the pollen grains, however, carry doubleness, and used on the female flowers of doubles give offspring all double (single being the dominant). The pollen of this plant is as uniform and perfect as that of any species I have ever seen. We must therefore conclude that the segregation by which singleness separates from doubleness is effected not later than the formation of the rudiments of the male and female flowers. Cytological investigation may no doubt show that the distinctions between the genetic properties of the male and female are associated with visible nuclear differences, but I see no reason for anticipating that such differences must exist. Cells which differ in their genetic potentialities must of course differ in physical constitution, but there is no reason to suppose that this difference need be in any way dependent on chromosome structure.

[9] *Jour. Gen.*, VIII, 1919, p. 199.

As regards *Campanula carpatica "pelviformis"* and *Begonia Davisii,* experiment has shown that the peculiar kind of segregation which they exhibit may recur in their offspring. In the *Begonia,* if the female of *Davisii* be fertilized with pollen of an ordinary double tuberous *Begonia,* the doubleness so introduced stays on the male side just as the doubleness of its own male does, and a plant so bred has its pollen all double. But if the male of *Davisii* be used on the female of an ordinary single, there is no restriction of doubleness to either sex of the offspring. The peculiarity of *Davisii* must therefore be attributed to the special properties of its female side. The *Campanula* case is complex and has not yet been fully explored, but at least from the female side of *pelviformis* plants have been raised which retain the properties of the mother as regards the distribution of the white and blue colors.

We have at the John Innes Institution been lately investigating a simliar case in flax, which, though comparable, has some special features. A dwarf flax (*Linum usitatissimum*) of unknown origin, presumably a stray seedling of one of the varieties grown for oil, was fertilized with pollen from our tall fiber strain. Both parents breed true to the fully hermaphrodite condition, with anthers perfectly formed, and the F_1 plants were normal in this respect. F_2 consisted of hermaphrodites, and a recessive form with aborted anthers, generally contabescent and not dehiscing at all, but having occasionally a few grains of good pollen. The ratio was a normal 3:1. The recessive, having occasional grains of pollen, self-fertilized, gave similar plants with anthers wholly or almost wholly aborted. The normal F_2 hermaphrodites gave in F_3 families which showed that some of the F_2 plants were pure normals, others heterozygous in the ordinary way. But when the recessives were fertilized with pollen from three several varieties of tall fiber flax, only recessives were produced. These tall flaxes therefore are normally heterozygous for the recessive or "subfemale" condition, and this in segregation is perman-

ently relegated to the male side of the plant, while the female side takes the hermaphrodite factor. Segregation in regard to the same recessive may take place in one of two ways. It may be *unilateral,* as it is when in heterozygous association with the female of the tall flaxes, or it may be *ambilateral* and unrestricted to either sex when it is in association with the female of the oil flax. We must infer that the female halves of the two types differ in some critical respect which decides the manner of the segregation.

Unilaterality may also show itself as a difference in the closeness of linkage on the two sex-sides of the same plant, and no doubt this fact may have a bearing on the interpretation of the foregoing cases. The late R. P. Gregory discovered the first case of this, in *Primula sinensis.* He found that the linkage between magenta color and short style was closer in the eggs than in the pollen. Recent work on a larger scale has given 10.9:1 as the female linkage and 6.4:1 for the pollen. A similar difference has been also found for the linkage between green stigma and "reddish" stem (as opposed to dark red), the value being 29.8:1 for the eggs and 41.7:1 for the pollen. In both examples, individual families show wide fluctuations, and these values should for the present be regarded as approximate only. Whatever be their meaning, they show that some segregation has occurred in the formation of the two sets of sexual organs, such that the process of gametic differentiation is not the same in both.

Besides these examples of differentiation between the male and female sides, there are others proving that segregation may occur at other stages in somatic development. The most obvious examples are the variegated plants. I have discussed this subject elsewhere in connection with reversible periclinal "chimæras" of white over green which produce shoots having the white inclosed with the green.[10] To these must now be added the cases in which the plants arising from adventitious

[10] *Jour. Gen.,* Vol. 7, p. 93 (1919).

buds differ from the plants which produce them. I have described one of these examples in *Bouvardia*. The pinkish white "Bridesmaid" gives the red-flowered "Hogarth" from its root-cuttings. Three similar occurrences have been found in fancy Pelargoniums. The root-cuttings of a white-flowered variety, "Pearl," give a red-flowered form very like "Mme. Thibaut." "Mrs. Gordon," which is a full rose-pink, with whitish edges, gives from its root-cuttings flowers in which the two posterior petals are marked with dark red, not unlike the variety "Cardiff." A more striking case is that of "Escot," which gives from its roots plants with bright pinkish red marks, those of the original being purplish red. The most curious feature of this case lies in the increased size both of the plant and the flowers coming from the roots, and it is scarcely possible to see the petals of "Escot," which are characteristically rolled back, side by side with those of the root-form, which are not only larger, but also flat, without surmising that this rolling back is an expression of the greater size of the larger petal contained within the smaller, causing a want of correlation between the growth of the inner and outer tissues.

Buckling or crumpling of leaves through want of correlation was a conspicuous feature of some of Winkler's "graft-hybrids," made from *Solanum nigrum* and *S. lycopersicum*, when the larger tomato was inclosed within the smaller species. We have had a precisely similar example in a salmon-fringed Pelargonium bred by Mr. Jarman, of Chard. The leaves are obviously buckled, the petals are laciniated, and the female parts aborted, though the anthers are perfect. This male and deformed flower is proper to the outer tissues only; for on two occasions the plants have produced shoots with large flat leaves and normal hermaphrodite flowers with their petals entire. Obviously, this normal plant was inclosed within a skin of the fringed type.

In all these examples, a somatic segregation has occurred which determines the genetic potentialities. The

interpretation that they are *periclinal* chimæras is probably correct for the most part. The fringed *Pelargonium* is obviously of this nature. Nevertheless, the fact that a root-cutting consistently produces a certain type of plant which is not the original does not prove that the distribution is periclinal. Another possibility is well illustrated by the case of a variegated *Spiræa ulmifolia*, having the stem, petioles, and (basal) centers of the leaves without chlorophyll.[11] The growing point has the power of laying down green tissue in the lateral areas only, the internodal regions being albinotic. Root-cuttings from this form give albino plants which die after the development of two or three small leaves. Now in this case we can see the distribution of the green and white, respectively, and we recognize that the roots give albino plants because they belong wholly to the albinotic area. On similar lines it is possible to interpret the *Bouvardia* and other cases. The distribution of the two types in the same plant may be such that one is limited to the root and internodes, while the other is in the nodal structures.

That considerations of this kind are not fantastical is proved by the genetical phenomena seen in the case of "rogues" in culinary peas, which Miss Pellew and I have been investigating for a number of years.[12] The rogue is a peculiar, wild-looking plant, differing in various ways from the type, chiefly in having pointed leaflets. Crosses between it and the type give plants which in their lower parts are intermediate, though turning into rogues as they develop. The self-fertilized offspring of rogues and also of these F_1 plants are always rogues, and evidently the type-characters introduced

[11] This is somewhat like the *Pelargonium* named by Messrs. Cannell "Freak of Nature," in which the chlorophyll has a closely similar distribution, and it, like the *Spiræa*, is sterile on both male and female sides. In this *Spiræa* I have never seen pollen, but very rarely a fruit is formed, which, no doubt, is due to an occasional development of a bud in the green area, an occurrence frequent in variegated plants. Whether these fruits contain viable seeds is not yet known.

[12] *Roy. Soc. Proc.*, B, Vol. 91, p. 186 (1920).

from the type parent are left behind in the lower parts. Such a case may perhaps be compared with the condition seen in the variegated *Spiræa,* and we may fairly conjecture that if it were possible to raise root-cuttings from the F_1 peas, they would produce types.

A more gradual exclusion of the type-elements in the lower parts is seen in certain intermediates. These may scarcely differ from types in the lower parts, though changing to rogues, sometimes abruptly, sometimes gradually, as the series of flowering nodes is developed. Reciprocal crosses between the successive flowers of such plants and the flowers of types has shown that, together with the gradational change in the somatic structures, there is also a gradational change in the proportion of gametes bearing the rogue and type characters respectively. This proportion and the rapidity of the change differ on the male and female sides. Of the *egg cells* in the lower flowers, up to about the tenth flowering node, rather more than 50 per cent. carry the type-characters —or at least the non-pointed leaflets—but above this level the proportion of types declines. Of the *pollen* in the lowest two flowers only about 20 per cent. is type-bearing and the proportion diminishes rapidly in each successive flower above the level.

In all the examples given hitherto the segregation is in diploid tissues, but a comparable phenomenon has been proved by Collins to occur in the *haploid* axis of a moss (*Funaria*). In a dicœcious moss, as the Marchals have shown, sex-segregation occurs at spore-formation, the division in which reduction is effected. This, of course, agrees with cytological expectation, though, so far as I know, the details have not been observed. But from the leaves of mosses placed in nutrient fluids new plants may be raised without great difficulty, and Collins found that the (perigonial) leaves surrounding the male organ thus propagated, produce *exclusively male* axes.[13] He has since raised similar cultures from the (peri-

[13] *Jour. Gen.,* Vol. 8, p. 145 (1918–19).

chætial) leaves surrounding the female organ, and, as related in his recent paper (Genetics of Sex in *Funaria hygrometer*), from them monœcious plants resulted. The proof is thus complete that in a haploid tissue a segregation of sex can occur.

The inference may be drawn that the factors for other characters may similarly be liable to segregate in the haploid state. In this connection I may mention a case which, though as yet obscure, perhaps fulfils this expectation. In botanic gardens a variegated maidenhair fern (*Adiantum capillus-Veneris*) is grown which has wedges of white tissue irregularly distributed in the segments. This plant produces spores freely,[14] and these give rise to prothallia which in several cultures raised here have always been entirely green. But when ferns arise from these green prothallia by the sexual process they are of three kinds, green, white, and variegated like the parent plants. The fact that the prothallia should be all green is entirely unexpected and creates a distinct problem, but it is evident that segregation must occur either in some of the cell-divisions by which the prothallia proliferate, or in those by which the gametes are formed; in either case in haploid tissue. This segregation is essentially different from that by which the differentiation of organs, such as the archegonia and antheridia, is accomplished, inasmuch as it relates to elements determining the characters of the next generation.

From the evidence given it is clear that in a wide view of living things segregation can not be exclusively a property of the reduction-division, and for the present it should be regarded as a possibility which may occur at any division in the life-cycle.

[14] I have not satisfied myself that spores are produced in sori on the white areas.

FACTORS IN THE RESISTANCE OF GUINEA PIGS TO TUBERCULOSIS, WITH ESPECIAL REGARD TO INBREEDING AND HEREDITY

DR. SEWALL WRIGHT, S.D., AND DR. PAUL A. LEWIS, M.D.

BUREAU OF ANIMAL INDUSTRY, U. S. DEPARTMENT OF AGRICULTURE, WASHINGTON, D. C., AND THE HENRY PHIPPS INSTITUTE, THE UNIVERSITY OF PENNSYLVANIA, PHILADELPHIA, PA.

IN the pre-bacteriologic era physicians were quite generally of the opinion that heredity played a large part in resistance or susceptibility to tuberculosis. Following Koch's demonstration of the part played by the *Bacillus tuberculosis* in the etiology of the disease it was recognized that the hereditary influences might be only apparent; the disease being established in any family there was evidently an immeasurably increased chance of interfamilial infection; and as a consequence of this uncertainty of interpretation it has become quite customary to regard hereditary influences, properly considered, as a negligible factor.

Pearson[1, 2] and Goring[3] compared the correlation between parent and offspring, in incidence of the disease, with that between husband and wife. The effects of unfavorable conditions and the chances of infection might be about as great in one case as in the other, while heredity would be a common factor only in the first case. Pearson dealt with upper class families of which one member was being treated in a certain sanitarium. The correlation between parent and offspring came out about .50, in the usual scale, in which 1.00 is perfect correla-

[1] Pearson, Karl, 1907, "A First Study of the Statistics of Pulmonary Tuberculosis." Dulau & Co., London. 26 pages.

[2] Pearson, Karl, 1912, "Tuberculosis, Heredity and Environment." Dulau & Co., London. 46 pages.

[3] Goring, Charles, 1909, "On the Inheritance of the Diatheses of Phthisis and Insanity." Dulau & Co., London. 28 pages.

tion, after making certain assumptions in regard to the frequency of the disease in the whole population, and making allowance for the age of the children, many of whom had not reached the age of greatest danger. This correlation is about the same as that between parent and offspring in characters such as height, weight, eye color, etc., which are largely or wholly determined by heredity. The correlation between husband and wife was only .24 and even much of this, the author found reason for attributing to assortative marriage.

Goring's study was made with the families of British convicts. He also found a correlation of about .50 between parent and offspring but no correlation (— .01) between husband and wife. The only assumption that had to be made in this case was a correction for the age of the children.

We are acquainted with no experimental work bearing on this question.

Material

The present paper describes the results of experiments on certain inbred and crossbred stocks of guinea pigs. All of the animals used were a by-product of experiments on the effects of inbreeding which have been carried on by the Bureau of Animal Industry of the U. S. Department of Agriculture since 1906. One of the authors (Wright) has been in charge of these experiments since 1915. A detailed report on the results is soon to be published. A brief summary will suffice here.

Twenty-three families of guinea pigs were maintained for a number of years by mating exclusively brother with sister among the descendants of twenty-three original pairs. In sixteen cases (including families 2 and 13 of those tested for resistance to tuberculosis) both of the original parents came from a stock which had already been maintained for twelve years without the infusion of fresh blood, by the Experiment Station of the Bureau of Animal Industry. In the remaining families, including families 32, 35 and 39, the original females came from

the above mentioned stock, while the original males were purchased from a local dealer.

In 1911 a number of animals were selected from the stock of the Experiment Station to start a control experiment. The mating of even second cousins has been avoided in this stock, which has been called Experiment *B*.

Eighteen of the inbred families were still on hand in 1916, having then on the average about eleven generations of brother-sister matings back of them. At this time most of the families were disposed of in order to make room for cross-breeding experiments and to obtain larger numbers from the five families which it was decided to retain. These families, numbers 2, 13, 32, 35 and 39, are the ones which have been tested for resistance to tuberculosis. They were retained in part because they already occupied many pens, but largely because of the possession of contrasting characteristics in size, fertility and coat pattern. Each of these families is at present very homogeneous in heredity. Family 2 is descended wholly from one mating in the sixth generation of inbreeding. Families 13, 32, 35 and 39 come from single matings in the seventh, eleventh, twelfth and eighth generations respectively.

The total number of animals involved in the experiment on inbreeding and cross-breeding has been about 30,000.

THE EFFECTS OF INBREEDING ON VIGOR

It is noteworthy that there has been no very obvious decline in vigor, although the families are now on the average in the fourteenth generation of brother-sister mating and one of the most vigorous (No. 35) has reached the twenty-first generation.

There has, however, been some decline in vigor in all respects which have been studied. The decline is most marked in fertility, including both frequency and size of litters. It has been so great in this respect that it would have to be recognized even though the decline in

other respects were assumed to be due wholly to less favorable environmental conditions.

That there has been a real genetic decline in the inbred stock in all elements of vigor is shown by comparison with the control stock *B*, which has been superior in every respect. Still better evidence has been obtained by comparison of the inbreds with the young from crosses between the different families raised at the same time and under the same conditions.

The Effects of Crossing

In interpreting the effects of crossing, the characteristics which depend on the hereditary make-up of the young must be distinguished from those which depend on the dam or sire. In studying these questions, inbred females have been mated with inbred males of another family (experiment *CO*) and with crossbred males (experiment *CA*). Crossbred females have been mated with brothers (experiment *CI*), unrelated crossbred males (experiment *CC*) and inbred males of an unrelated family (experiment *AC*).

Size of litter appears to depend wholly on the dam. There is little or no improvement in the experiments in which the dam is inbred (*CO, CA*). There is, however, a marked increase, 10–30 per cent. depending on conditions, in the litters produced by crossbred females (*CI, CC, AC*).

The record of an experiment in frequency of litters depends on the age of maturity as well as on the regularity thereafter. Males mature considerably later than females, so that the age of maturity of the male is the controlling factor in this respect in matings, made as in the present experiments, between immature animals. The frequency of litters after maturity appears to depend largely on the dam. There is no improvement in the record of the first cross (*CO*) over the inbreds. There is, however, marked improvement in the other experiments in which either the sire or dam or both are cross-

bred. Crossbred males mature earlier and the crossbred females have litters more regularly than inbreds.

The percentage of the young born alive depends almost wholly on the dam. There is little or no improvement in experiments *CO* and *CA*, but an increase of 6 to 8 per cent. where the dam is crossbred. The percentage which are raised to 33 days of age of the young born alive depends both on the dam and on the heredity of the young. There is a marked increase, 9 to 12 per cent., in all of the crossbreeding experiments mentioned above.

Somewhat similarly, birthweight depends largely on the dam, while the gains between birth and 33 days depend to a considerable extent, though far from wholly, on the heredity of the young. Guinea pigs become independent of the dam at a very early age. There is an increase of 2 or 3 per cent. in experiments *CO* and *CA*, but one of about 7 to 10 per cent. where the dam as well as the young are crossbred. In the gain between birth and 33 days, there is an improvement of about 16 per cent. in the first cross, which is somewhat increased in the young produced by crossbred dams. There is an increase of 15 to 20 per cent. in the adult weight in the first cross (*CO*). This is maintained in the young produced by matings of unrelated crossbreds (*CC*), but half of it is lost where the parents, though crossbred, are brother and sister (*CI*). The influence of the dam does not appear to extend to the adult weight.

A loss in the improvement brought about by the crossing becomes apparent in the second generation of inbreeding following a cross (experiment *C*2) in those cases in which it is not apparent in the first generation.

COMPARISON OF DIFFERENT FAMILIES

A comparison of the different inbred families with each other has revealed persistent differences in color, pattern, tendency toward polydactylism, tendency toward production of monsters, mortality among the young, weight and both elements of fertility. It was found that

the differences in these respects could not be interpreted merely as differences in general vigor. Vigor above the average in one respect was as likely as not to be found associated with a subnormal record in another respect, the correlation between the records of the families in two respects coming out in most cases substantially zero. Examples of these family differences will be brought out later in connection with the effects of inoculation with tuberculosis.

Explanation of the Results of Inbreeding and Crossing

These results harmonize well, on the whole, with those found by other investigators. It is believed that they can be explained as consequences of the current theory of heredity without recourse to the rather mystical ideals which once prevailed in regard to inbreeding. There appear to be independently inherited factors which affect frequency and size of litter, ability to bear the young successfully, vitality and growth as well as for color, pattern and the other characters in which the families differ. There seem to be surprisingly few factors which act on all of these characteristics. The concept, hereditary vigor, thus becomes merely an expression for the sum of a number of independently inherited qualities and not an entity.

The factors which cause reduced vigor in any respect appear to be in general recessive. The primary effect of inbreeding is to render homozygous a random group of the factors present in the original stock. Some combination of factors, good, bad and indifferent, thus becomes fixed in each inbred line. As the recessive factors, tending toward lack of vigor, are as likely to become fixed as the dominant ones there is on the average a decline in vigor in each respect. Moreover, owing to the likelihood that many factors for vigor will be linked genetically with factors causing weakness, it is to be expected that vigor in all respects will be found in very few lines, even where there is careful selection. In the pres-

ent case, there was very little conscious selection but a considerable amount of natural selection was, of course, unavoidable.

On crossing two inbred lines, each, as a rule, supplies some of the dominant factors lacking in the other, with the consequence that there is increased vigor in so far as the character in question depends on the heredity of the animal itself. In the next generation if brother-sister matings are made, there should be a decline as compared with the first generation in characters which depend wholly on the animal itself. The decline from this cause may, however, be balanced or more than balanced by the improvement due to the influence of the crossbred dam.

EXPERIMENTS ON RESISTANCE TO TUBERCULOSIS

Since April, 1919, all surplus individuals in five inbred families (2, 13, 32, 35 and 39) and a random selection from certain crossbreeding experiments (*CO, CA, AC,* and *B*) have been shipped from Washington to Philadelphia to be tested for resistance to tuberculosis. The animals were shipped in lots of 30 to 60 every two or three weeks. They were from 5 to 8 weeks old at the time of shipping. They usually arrived in good condition but with some loss in weight. The first two lots were not tested because of heavy mortality before inoculation due to feed conditions. Lot 6 was not tested because of heavy mortality due to excessive heat during shipment. Data on the shipment and inoculation are presented in Table I. The deaths within 15 days after inoculation are probably not attributable to tuberculosis and are not considered in the later work. The experiments were considered closed when all but two or three of the animals had died. The last column gives the number of days after inoculation at this time. The next to the last column gives the day by which 50 per cent. of those which passed 15 days had died. It agrees fairly well with the average number of days calculated on the assumption that the few survivors of the experiment died on the day following that given in the last column.

The cultures used were of the human type of tuberculosis with the exception of lot 3 in which the type was bovine.

TABLE I

Lot	Date Shipped	Date Inoculated	Inoculation	No. Died Before 15 Days	No. Survived 15 Days	Av. No. of Days Lived	Day to which 50 % Survived	Day Exp. Closed
3	May 4	June 2, 1919	1/10 mg. culture 07.8 intraperitoneal	2	32	43.9	44	62
4	June 4	" 10, "	1/5 " 465 "	5	30	25.9	21	40
5	" 18	July 3, "	1/10 " 465 "	7	57	24.0	24	31
7	July 17	Aug. 25 "	1/5 " DG "	3	37	25.6	24	43
8	" 30	" "	" " " "	1	28	25.2	24	38
9	Aug. 14	" "	" " " "	3	44	25.3	24	39
10	" 28	Oct. 24 "	" " " subcutaneous	6	46	49.7	47	84
11	Sept. 11	" 25 "	" " " "	3	31	54.2	54	83
12	" 25	" " "	" " " "	2	30	49.5	46	83
13	Oct. 9	" " "	" " " "	3	42	45.2	40	83
14	" 23	" " "	" " " "	2	35	42.3	39	83

Lots 7, 8 and 9 were given the same inoculation on the same day. The results are so similar, in spite of the differences in the age and weight of the animals, that they can be dealt with satisfactorily as one experiment. Lots 10 to 14 were all given the same inoculation on October 24 and 25 and can also be dealt with as one experiment. The same quantity of the culture, inoculated intraperitoneally in lots 7 to 9, was inoculated subcutaneously in lots 10 to 14, with the consequence that the average length of life was much greater in the latter group of lots. Lots 3, 4, and 5 were inoculated separately with different cultures or different amounts of the same culture. The numbers in each case are rather small for separate treatment. The mortality curves for lots 4 and 5 are not, however, very different. That for lot 3 can be made similar by multiplying the days of survival after inoculation by 3/5. These three lots have been combined in this way for comparison with the later and more satisfactory experiments.

SEX AS A FACTOR IN RESISTANCE

In attempting to analyze the difference in the length of life following inoculation, we will consider first a number of possible factors other than heredity. It will be convenient to start with sex. Chart 1 shows the per-

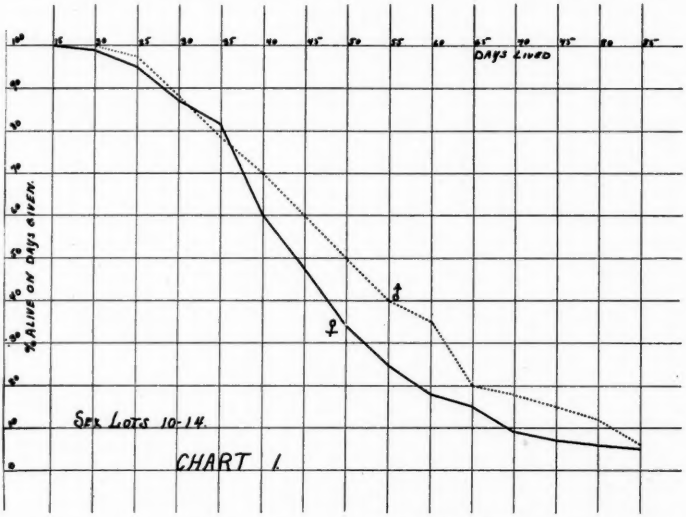

CHART 1. The mortality curves of the males and females of lots 10 to 14. This and the later charts give the percentage alive at the beginning of each five-day period starting with 100 per cent. on the fifteenth day after inoculation.

centage of the males and females alive at the beginning of each day in lots 10 to 14, starting with 100 per cent. on the fifteenth day after inoculation. The males have some advantage in lots 10–14, but it is too small to be of more than doubtful significance. In lots 3 to 9, moreover, the two mortality curves keep crossing each other in such a way as to indicate that sex makes no appreciable difference in resistance.

THE AGE OF THE DAM AS A FACTOR

Some rather dubious indications of a relation between age of mother and susceptibility of the offspring to tuberculosis were found by Pearson among human beings.

The guinea pigs in lots 10–14 were divided into two groups, those containing at least 25 per cent. of the blood of family 35 and the remainder. Each of these groups was classified by birth rank with results given in Table II.

TABLE II

AVERAGE NUMBER OF DAYS SURVIVAL AFTER INOCULATION AMONG THE YOUNG IN DIFFERENT BIRTH RANKS, LOTS 10–14

Order of Litter	Animals with Blood of Family		Miscellaneous	
	No.	Av.	No.	Av.
1	16	58.9	17	35.6
2	21	60.3	29	43.1
3	7	70.6	23	43.3
4	12	63.3	26	39.8
5	4	60.8	6	35.8
6			11	45.9
7			5	37 1
8			3	33.3
8 +			4	30.8

These figures suggest an increase in resistance of the young born in litters up to the third and a decline in resistance in young born of aged dams. They must be confirmed by larger numbers, however, before much significance can be attached to them.

THE RELATION OF AGE, WEIGHT AND RATE OF GAIN TO RESISTANCE

Mere bulk must be considered as likely to be a factor in resistance to tuberculosis. Other things being equal, one would expect to find that the same inoculation given to a St. Bernard and to a toy spaniel would have a more disastrous effect on the latter. In lots 10–14, some of the guinea pigs were over three times as heavy as others at the time of inoculation (variations between 120 and 440 grams).

The rate of gain is a good indication of the condition of the animals and is thus another factor which one would expect to find correlated with resistance to disease. In this connection, we have for study the birth-weight, weight at 33 days and the rate of gain between 33 days

and inoculation. The birthweight varies between 40 and 120 grams, the weight at 33 days between 90 and 360 grams, and as to rate of gain after 33 days, some lost weight while others gained over 3 grams a day.

Age is a factor which must be considered apart from weight and rate of gain. In lots 10–14, there were varia-

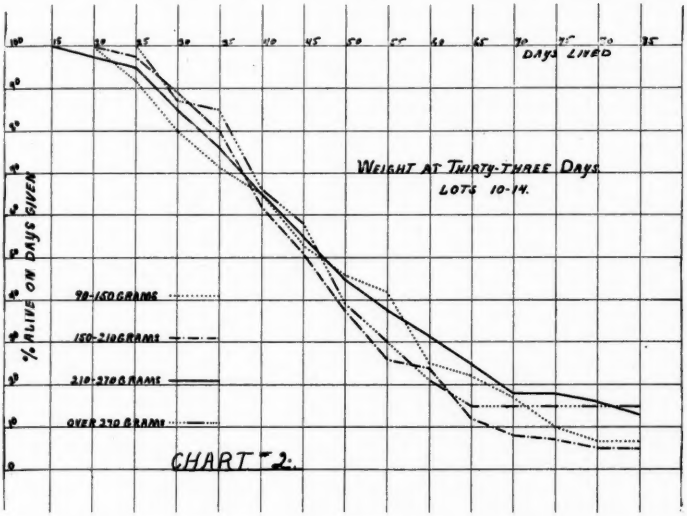

CHART 2. The mortality curves of the guinea-pigs of lots 10–14, grouped according to weight at weaning.

tions between 35 and 105 days at the time of inoculation. In lots 7–9 the variation was between 45 and 85 days.

The correlation between the length of life after inoculation and each of these factors, including also size of litter, is given in Table III for lots 7–9 and lots 10–14. All of these correlations were calculated by the usual product

TABLE III

Factors Correlated	Lots 7, 8, 9	Lots 10–14
Days lived—Size of litter	+ .090 ± .064	+ .029 ± .050
—Birth weight	— .044 ± .064	+ .059 ± .050
—Weight at 33 days	+ .022 ± .065	+ .056 ± .050
—Rate of gain (33-inoc.)	+ .120 ± .064	+ .223 ± .047
—Age at inoculation	+ .025 ± .065	+ .180 ± .048
—Weight at inoculation	+ .092 ± .064	+ .219 ± .047

moment method. Perfect positive correlation is +1, perfect negative correlation is −1, while 0 indicates the absence of correlation.

Inspection of these figures reveals that all of them are surprisingly low. In lots 7–9, the factor most closely correlated with length of life is the rate of gain between weaning (33 days) and inoculation, and even this is not certainly significant in view of its probable error (+.120 ± .064). Larger correlations are to be expected in lots 10–14, owing to the greater heterogeneity in age and weight, but even here the correlations are all small. Size of litter, birthweight and weight at 33 days are again of no value as indicators of resistance to tuberculosis. There, are, however, significant correlations between length of life and rate of gain (+.223 ± .047), age (+.180 ± .048) and weight of inoculation (+.219 ± .047).

The degree to which variation is determined by a given factor is measured by the square of the coefficient of correlation. On this basis, only 5 per cent. of the variation in length of life (*i.e.*, 5 per cent. of its mean square deviation) is determined by the most important of the above factors. The degree of determination by all of these factors combined can not be found by merely adding the squares of the correlations, because the factors are not independent of each other. For example, rate of gain is an element in determining the weight at inoculation. Any effect which it has on resistance should be reflected in a correlation between weight and length of life as well as between rate of gain and length of life. The correlations among the more important of these factors are given in Table IV.

TABLE IV

		Lots 7, 8, 9	Lots 10–14
Weight at inoculation	—Rate of gain.....	+ .357 ± .056	+ .553 ± .035
	—Age	+ .420 ± .054	+ .742 ± .022
	—Weight at 33 days.	+ .780 ± .025	+ .670 ± .027
Rate of gain	—Age	+ .323 ± .058	+ .592 ± .032
	—Weight at 33 days.	— .153 ± .063	— .040 ± .050
Weight at 33 days	—Age	— .012 ± .065	+ .160 ± .048

Weight at inoculation is of course completely determined by the weight at 33 days and the rate of gain and age at inoculation. The correlations between weight at inoculation and these three factors are accordingly high. They are rather different in the two groups of experiments, but this is to be expected. There was much less

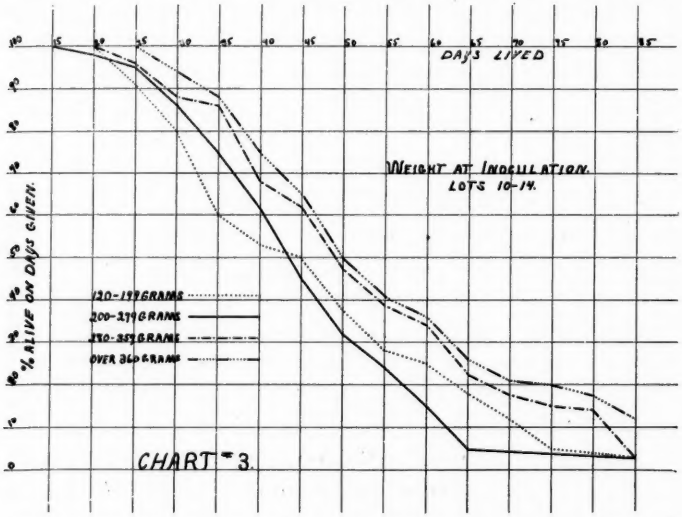

CHART 3. The mortality curves of the guinea-pigs of lots 10–14, grouped according to weight at inoculation.

variation in age in lots 7–9 than in lots 10–14 and variation in age accordingly plays a less important part in determining the variations in weight than in the latter case. The rather high correlation between age and rate of gain is due to the fact that nearly all of the guinea pigs suffered a loss in weight following shipment at a little over 33 days of age. Thus the older they were at inoculation, the greater the time in which they had to recover from the effects of shipping. The rate of gain before 33 days is generally an indication of the probable later rate of growth. In the present case, however, the slight negative correlation between the rates of gain before and after 33 days seems to indicate that shipping delayed

more the growth of those guinea pigs which had been gaining rapidly than of those which had more growth to make up. The small positive correlation between weight at 33 days and age at inoculation reflects a decline in the condition of the stock, due probably to the use of an inferior quality of hay, which took place during the sum-

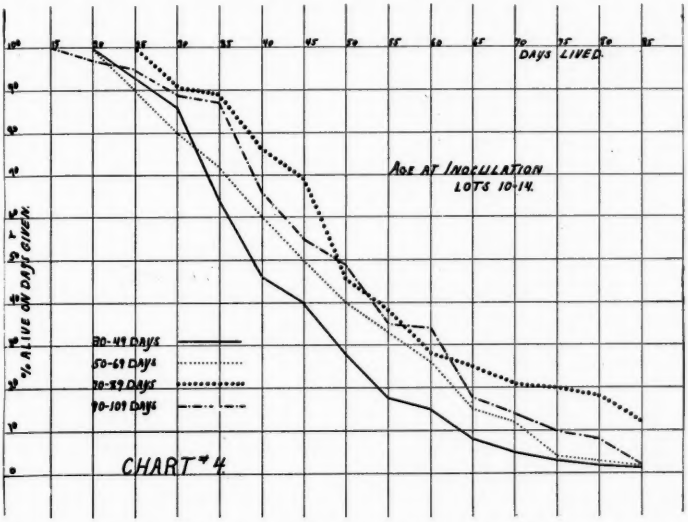

CHART 4. The mortality curves of the guinea-pigs of lots 10–14, grouped according to age at inoculation.

mer of 1919, especially during the time in which lots 10–14 were being raised.

With the help of these correlations, we can calculate the degree to which variation in length of life is determined by each factor separately and by all of them combined. For the last purpose, Pearson's coefficient of multiple correlation can be used. This coefficient comes out + .136 in lots 7–9 and + .251 in lots 10–14 for the correlation between length of life and rate of gain after 33 days, age and weight at inoculation combined. The degree of determination is measured by the square of this coefficient. These three most important factors combined, therefore, determine less than 2 per cent. of

the variation in length of life in lots 7–9 and less than 7 per cent. in lots 10–14. The separate contributions of the various factors can be calculated with results given in the following table.

TABLE V

DEGREE OF DETERMINATION OF DAYS LIVED AFTER INOCULATION BY AGE, WEIGHT AND RATE OF GAIN

Factors	Lots 7–9	Lots 10–14
Direct effect of age	.0015	.0005
weight	.0049	.0229
rate of gain	.0116	.0233
age and weight	—.0023	—.0051
age and rate of gain	—.0027	—.0041
weight and rate of gain.	+.0054	+.0256
Total degree of determination	.0184	.0631

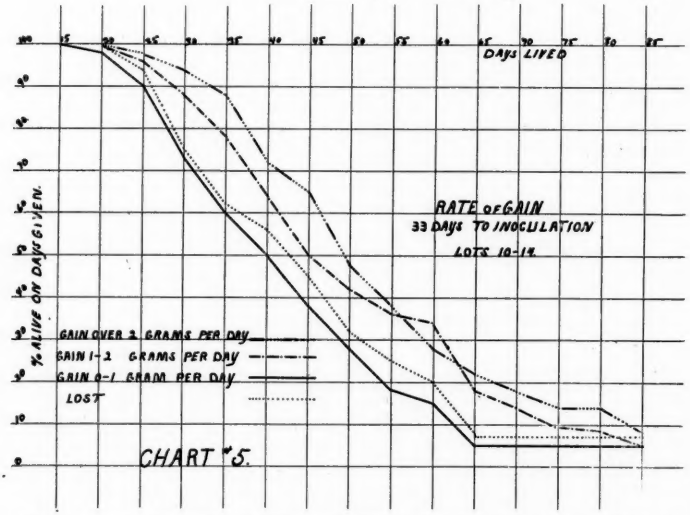

CHART 5. The mortality curves of the guinea-pigs of lots 10–14, grouped according to the rate of gain between weaning and inoculation.

While these figures can not be trusted in detail, owing to the large size of the probable errors of the correlations on which they are based, they indicate that age within the limits of one to three months has virtually no direct effect whatever on resistance to tuberculosis. There seems to be a slight direct effect of absolute weight

and a slightly greater direct effect of rate of gain, presumably as a measure of the condition of health.

NOTE

It may be of interest to readers who are not familiar with this method of analysis to compare the degrees of determination shown in Table V with those in a case in which we know a priori that there is complete determination of one variable by a number of others. The data for such a case have already been presented. The weight at inoculation is completely determined by weight at 33 days and the product of the rate of gain and interval between 33 days and inoculation. The coefficient of multiple correlation between weight at inoculation and these three factors combined comes out .9884 in lots 7–9 and .9628 in lots 10–14. The degrees of determination are as follows:

TABLE VI

Factors	Lots 7–9	Lots 10–14
Direct effect of weight at 33 days	.7106	.3689
rate of gain	.1501	.0907
age	.0930	.2177
Joint effect of weight at 33 days and rate of gain.	—.0500	—.0073
weight at 33 days and age	—.0031	.0907
rate of gain and age	.0763	.1663
	.9769	.9270

The departure of the multiple correlations and the sums of the degrees of determination from unity are of the order usually met in such cases. This method of analysis applies strictly only where the effects of the factors are combined by addition and the correlations are linear, conditions which are not perfectly met in the present case.

The facts brought out by the method of correlation are presented graphically in Charts 2, 3, 4 and 5. These charts show the decline in numbers, on the basis of 100 per cent. alive on the 15th day after inoculation among groups of guinea pigs of lots 10–14, classified by weight at 33 days, weight at inoculation, and rate of gain.

As a result of the foregoing considerations, it must be concluded that the apparent condition of a guinea pig at the time of inoculation and a knowledge of its past history give exceedingly little indication as to its probable length of life after inoculation. Over 98 per cent. of the variation in length of life in lots 7–9 and over 93 per cent. in lots 10–14 is caused by factors other than those discussed. This leads us to a consideration of hered-

itary differences as one of the possible causes of this variation.

HEREDITARY DIFFERENCES IN RESISTANCE

The average length of life in each inbred family and crossbreeding experiment in lots 3–5, 7–9 and 10–14 is given in Tables VII. VIII and IX together with other

TABLE VII

AVERAGES OF THE CHARACTERISTICS OF THE GUINEA PIGS TESTED FOR RESISTANCE TO TUBERCULOSIS IN LOTS 3, 4 AND 5.

The days lived in lot 3 are multiplied by 3/5 to make the average about the same as in lots 4 and 5.

Family or Experiment	Number Tested	Size of Litter	Birth Weight	Wt. at 33 Days	Wt. at Inoculation	Age at Inoculation	Av. Days Lived
2.....	28	2.85	73.3	201.1	209.8	54.6	23.1
13.....	15	3.13	83.4	232.7	227.7	49.7	20.3
32.....	2	2.50	80.5	256.0	215.0	53.0	21.0
35.....	9	2.56	86.5	252.0	236.7	50.5	25.7
39.....	1	4.00	77.0	157.0	140.0	49.0	16.0
CO.....	10	2.70	82.1	236.5	243.5	51.5	26.0
CA.....	24	3.08	85.5	247.4	247.7	52.0	25.7
AC.....	21	3.76	77.8	230.2	223.6	51.3	27.4
B.....	9	2.33	101.2	277.9	280.6	54.2	25.8
Inbred.....	55	2.89	78.5	219.2	218.0	52.4	22.6
Crossbred....	64	3.14	84.6	244.4	243.7	52.0	26.3

TABLE VIII

AVERAGES OF THE CHARACTERISTICS OF THE GUINEA PIGS TESTED FOR RESISTANCE TO TUBERCULOSIS IN LOTS 7, 8 AND 9

Lots 7, 8, 9	No.	Size of Litter	Birth Weight	Weight at 33 Days	Weight at Inoculation	Age at Inoculation	Av. Days Lived
2.....	13	2.46	80.2	197.1	243.1	64.2	22.8
13.....	16	2.81	91.0	257.1	308.1	63.6	23.9
32.....	10	2.40	86.6	224.4	273.0	71.8	20.6
35.....	9	2.56	85.0	242.5	282.2	61.2	27.1
CO.....	13	3.00	80.2	226.2	286.2	67.8	31.5
CA.....	14	2.93	80.1	213.1	251.4	58.1	24.0
AC.....	21	3.38	74.6	213.0	257.6	61.9	25.6
B.....	13	3.38	83.7	249.5	320.0	69.2	24.8
Inbred.....	48	2.58	86.0	231.3	278.3	65.0	23.5
Crossbred....	61	3.20	79.0	223.6	275.6	63.8	26.8

data. The distribution of the deaths is given in Tables X, XI and XII. As already stated, most of the lots were brought to a close while a few animals were still living. This introduces an element of uncertainty into the aver-

ages, but the assumption that these survivors died on the following day is unfair only to the superior stocks.

TABLE IX

AVERAGES OF THE CHARACTERISTICS OF THE GUINEA PIGS TESTED FOR RESISTANCE TO TUBERCULOSIS IN LOTS 10–14

Lots 10–14	No.	Size of Litter	Birth Weight	Weight at 33 Days	Weight at Inoculation	Age at Inoculation	Days Lived
2.....	27	2.56	73.1	175.0	254.4	76.9	45.3
13.....	18	2.67	84.2	221.1	301.1	91.3	35.7
32.....	5	2.20	88.0	223.2	228.0	63.0	35.4
35.....	22	2.73	83.8	231.6	301.4	72.8	58.5
CO.....	26	2.81	82.8	210.9	274.6	66.8	52.7
CA.....	33	3.45	72.3	195.6	274.8	69.8	56.5
AC.....	31	3.26	75.3	213.7	273.9	68.1	45.9
B.....	22	3.09	79.5	220.1	280.9	67.7	40.0
Inbred.....	72	2.61	80.2	207.2	278.6	73.3	46.2
Crossbred....	112	3.18	77.0	209.0	275.7	68.2	49.4

TABLE X

THE NUMBER OF DAYS FROM INOCULATION TO DEATH IN EACH EXPERIMENT IN LOTS 3, 4 AND 5

The intervals in lot 3 are multiplied by 3/5 to make them equal on the average to those in lots 4 and 5. The deaths before 15 days are given in Table I. The animals in experiments *CO*, *CA* and *AC* are also classified by the amount of blood of family 35 at the bottom of the table. The last column includes two animals in lot 3 which survived at 61 days, one in lot 4 which survived at 42 days, and 3 in lot 5 which survived at 30 days.

	Experiment																
	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	30+	Total
2....	1	2	1			4	5		2	1	2	1	2	5	2			28
13....			2		2	3	5	1	2									15
32....						1		1										2
35....					1			2		1			1	1	2		1	9
39....		1																1
B.....						1	1	1			2	2		1			1	9
CO...							1	2	1	1	3						2	10
CA...				1	3	1	3	1		1	1	1	2	2	3		5	24
AC...					2	1	1			1	1	4	1	3	2		5	21
0 (35).					5	2	5	1	1	3	5	2	1	3	3		6	37
¼ (35).								1				1	1	1	1		1	6
½ (35).				1				1				2	1	1	1		5	12

THE EFFECT OF INBREEDING

In each case, the crossbreds (*CO*, *CA*, and *B*) lived a little longer on the average than the combined inbred families. This result is in line with the results of cross-

TABLE XI

THE NUMBER OF DAYS FROM INOCULATION TO DEATH IN EACH EXPERIMENT IN LOTS 7, 8 AND 9

	Experiment																									
	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40	41	42	42+	Total
2.....		3	1		1		3	2	2											1								13
13.....				1	2	4	1	4		2				1	1													16
32.....	1	1	2	2	1			3																				10
35.....						1		2	3							1	1			1								9
B.....						1	1	5	4	1																1		13
CO.....							3		2		1					1		1					2	1		1	1	13
CA.....						2	3	2	7																			14
AC.....			1	2	2	3	2	3	1				1	1	2		1			1		1						21
0 (35).....			1	2		4	6	1	5		1		1		2		1											24
¼ (35).....					1	1	2	2	2					1	1													10
½ (35).....					1			2	3								1			1		3	1			1	1	14

TABLE XII

THE NUMBER OF DAYS FROM INOCULATION TO DEATH IN EACH EXPERIMENT IN LOTS 10 TO 14

	Experiment															
	15	20	25	30	35	40	45	50	55	60	65	70	75	80	82+	Total
2.....		3	3	2	4	3	2	1	2	3	1	1	1	1		27
13.....			4	6	4	1	1	1	1							18
32.....	1			1	1	1	1									5
35.....				1	3	2	2	2	2	2	1	1	1	2	3	22
B.....		1	4	1	6	4	2	2	1	1						22
CO.....		1	1	1	3	3	3	3	3	2	1	2			3	26
CA.....				2	2	1	6	4	3	8	2		1	2	2	33
AC.....		2	4	2	4	3	6	3		2	2			2	1	31
0 (35).....		3	5	4	8	5	10	6	4	6					1	52
¼ (35).....				1	1	1	3	1	1	1	1			1	2	13
½ (35).....						1	2	3	1	5	4	2	1	3	3	25

ing on vigor in other respects. In the present case, however, the difference does not appear very striking. In particular, the superiority of the control stock *B* over the inbreds seems very dubious. An earlier test of this question was made by Mr. E. H. Riley and Dr. E. C. Schroeder, of the Bureau of Animal Industry, when the inbred stock was in the sixth and seventh generations of inbreeding. They found that the inbred was distinctly inferior to the control stock in resistance. At that time, however, eighteen families were on hand. It is not unlikely that the four families tested in the present experiment are a selected lot in respect to resistance.

Returning to Tables VII, VIII and IX, there are no differences between experiments *CO* (parents unrelated inbreds), *CA* (sire crossbred, dam inbred) and *AC* (sire inbred, dam crossbred), which indicate an influence of the breeding of the dam on the resistance of the young. There are rather large differences, it is true, but these are not consistent and must be attributed largely to heredity from particular families rather than to the system of breeding.

Differences in Resistance among Inbred Families

Passing now to a comparison of the different inbred families with each other, we come to results which appear much more striking than the differences between inbreds and crossbreds. In all three groups of experiments, one family, 35, stands out as distinctly more resistant than the others. It leads the average of the others by 16 per cent. in lots 3–5, 19 per cent. in lots 7–9 and 43 per cent. in lots 10–14. The more striking result in the last case is probably due to the weaker inoculation. In spite of the large amount of variation in each case, the probable errors put the superiority of Family 35 beyond question. Among the other families, family 2 is on the whole the most resistant. Families 13 and 32 are about equally susceptible. Only one animal from family 39 was tested. This one was one of the first in its lot, no. 7, to die, indicating low resistance in this family also so far as conclusions can be based on such a slender basis.

Inheritance of Resistance among Crossbreds

If there are hereditary differences in resistance, one might expect to find differences among the crossbreds depending on the families which went into their ancestry. A preliminary test of this point was made as follows: The length of life of each crossbred (*CO, CA* or *AC*) was entered under each of the four grandparental inbred families. Thus an animal in experiment *CA*, whose sire

was a cross between families 35 and 2 and whose dam was of family 13, was entered once under each of the former families and twice under family 13. The averages of the entries under each family in this tabulation are given in Table XIII. It will be seen that in each group of experiments, family 35 has a distinct lead over

TABLE XIII

AVERAGE NUMBER OF DAYS LIVED BY THE CROSSBREDS DESCENDED FROM EACH INBRED FAMILY

Each crossbred is entered under the family of each grandparent.

Family	Lots 3, 4, 5		Lots 7, 8, 9		Lots 10–14	
	No.	Av.	No.	Av.	No.	Av.
35	30	27.9	38	30.3	63	64.4
2	65	25.9	56	26.8	116	51.4
13	45	24.8	49	26.0	72	48.7
32	33	24.4	21	23.9	51	49.0
39	28	26.0	16	24.9	39	48.0
Misc.	19	26.0	12	24.2	19	48.8

the others as ancestral to resistant crossbreds. Family 39 and the other families which entered into the ancestry of the crossbreds appear to rank with the more susceptible families 13 to 32. It will be seen that the rank of families 35, 2, 13 and 32 as ancestors of resistant crossbreds is the same as their own rank in resistance. These results are brought out more clearly in a tabulation in which all of the crossbreds are classified as half-blood, quarter-blood and zero-blood of family 35. The last class may be divided into half-blood, quarter-blood and zero-blood of family 2. The results in comparison with those for family 35, 2 and the others combined are given in table XIV.

The half-bloods of family 35 are distinctly superior to family 35 itself, and thus much superior to their other ancestral families.[4] In the three groups of experiments

[4] The preliminary study of another large series of animals (lots 15 to 21) shows that while the order among the crossbreds, as related to the presence of the blood of family 35 has been maintained, the advantage of the half-bloods over family 35 is absent, the curves being almost the same with the half-bloods slightly less. The order of the inbred families it may be added remains exactly as described.

TABLE XIV

THE AVERAGE NUMBER OF DAYS LIVED BY THE CROSSBREDS WHICH ARE ½ BLOOD, ¼ BLOOD AND HAVE NO BLOOD OF FAMILY 35, THE LAST CLASS BEING CLASSIFIED SIMILARLY WITH RESPECT TO FAMILY 2

The averages for Families 35, 2 and the other inbred families combined are given for comparison.

	Lots 3, 4, 5		Lots 7, 8, 9		Lots 10–14	
	No.	Av.	No.	Av.	No.	Av.
½ blood (35)	12	28.9	14	32.2	25	65.6
¼ blood (35)	6	28.7	10	24.8	13	56.5
No blood (35)	37	25.0	24	24.4	52	43.9
No (½ blood 2)	16	25.9	11	24.4	32	46.0
blood (¼ blood 2)	16	24.4	10	24.0	13	39.2
(35) (No blood 2)	5	23.8	3	25.7	7	43.6
Family (35)	9	25.7	9	27.1	22	58.5
Family (2)	28	23.1	13	22.8	27	45.3
Other Inbreds	18	20.4	26	22.6	23	35.6

3–5, 7–9 and 10–14, these half bloods exceed family 35 by 13 per cent., 19 per cent. and 12 per cent. respectively in duration of life, and exceed the other inbred families by 31 per cent., 42 per cent. and 60 per cent. respectively. We have here more decisive evidence of the improvement in vigor in this respect which may follow a cross, than in the comparison previously made between the total inbreds and total crossbreds. The most probable explanation is the same as that applied to the improvement in fertility, weight and death rate following crosses, viz., that each inbred family tends to supply dominant factors, favoring vigor, which are lacking in the other. Applied to the present case, this means not only that family 35 possesses dominant factors for resistance, lacking in the other families, but that some or all of the latter may possess such factors lacking in family 35. By inbreeding among the crossbreds, it should be possible to develop a strain even more resistant than family 35 provided that the linkage relations do not interfere with the fixation of the factors for resistance in one strain.

The quarter-bloods of family 35 confirm the conclusions derived from consideration of the half bloods. They are about intermediate between the half-bloods

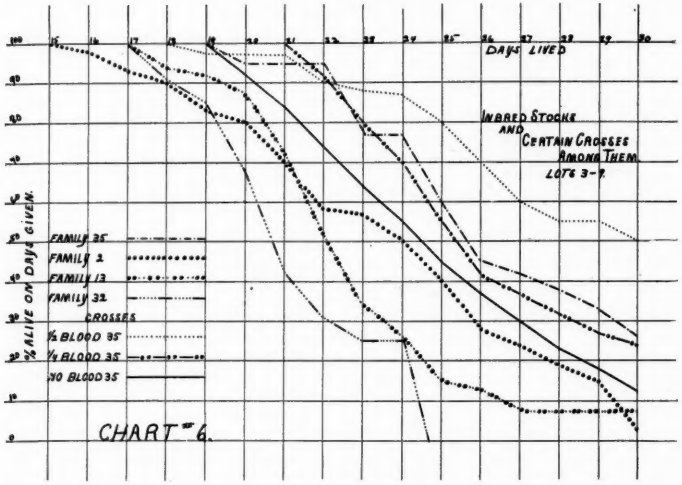

CHART 6. The mortality curves of the guinea-pigs of lots 3-9, grouped by breeding. Four inbred families. The crossbreds are grouped according to the amount of blood of the most resistant inbred family, No. 35.

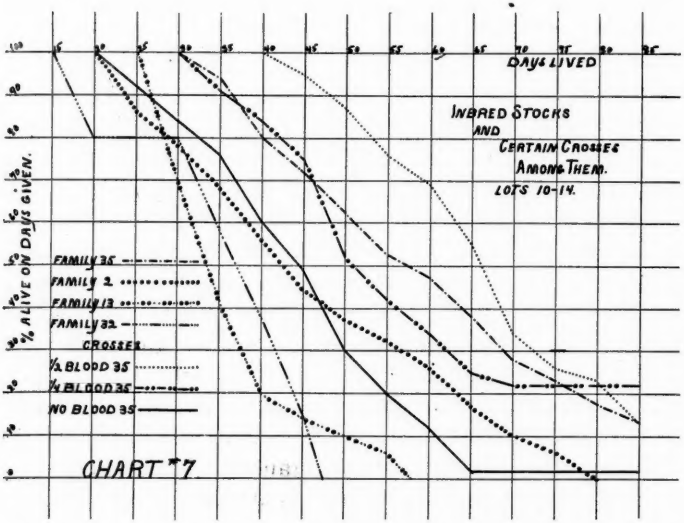

CHART 7. The mortality curves of the guinea-pigs of lots 10-14, grouped by breeding as in Chart 6.

and zero-bloods in lots 10–14 and in a combination of lots 3, 4, 5 with lots 7, 8, 9.

Curiously enough the crossbreds with no blood of family 35 do not show the improvement over their ancestral inbred families shown by the half and quarter bloods. They exceed the average of families 2, 13 and 32 by only 13 per cent. in lots 3–5, 8 per cent. in lots 7–9 and 7 per cent. in lots 10–14. They are not consistently superior to the best of these families, family 2. There seems to be dominance of resistance on the whole but no supplying of complementary factors for resistance such as was indicated in the crosses of family 35. This indicates that families 2, 13 and 32 are in the main susceptible because of the same genetic factors. The relations of the factors present in the different families is a question on which it is hoped more extensive evidence can be presented later (Charts 6 and 7).

Inheritance of Resistance and Sex

The crosses involving family 35 give some evidence on the inheritance of resistance from sire and dam. The average length of life in reciprocal crosses is given in Table XV.

TABLE XV

The Average Length of Life of Males and Females from Reciprocal Crosses Involving Family 35

	No.	Av. Days
♂ (35) × ♀ (misc.)	7♂	63.6
	5♀	59.4
	12	61.9
♂ (35 × misc.) × ♀ (misc.)	6	64.0
Total, (35) on sire's side	18	62.6
	No.	Av. Days
♂ (misc.) × ♀ (35)	7♂	73.6
	6♀	63.7
	13	69.0
♂ (misc.) × ♀ (35 × misc.)	7	50.1
Total, (35) on dam's side	20	62.4

The results are irregular, as might be expected with such small numbers, but they show that there is trans-

mission of resistance from each parent to offspring of each sex. The total figures—62.6 days where family 35 is on the sire's side, 62.4 where on the dam's side—indicate that there is probably equal inheritance from each sex.

DEGREE OF DETERMINATION OF RESISTANCE BY HEREDITY

We have shown that the length of life after inoculation was determined less than 2 per cent, by weight, age and rate of gain combined, in lots 7–9 and less than 7 per cent. by these factors in lots 10–14. It is interesting to compare the degree of determination by heredity with these figures. Such a comparison can be made for the crossbreds by finding the correlation between length of life and the amount of blood of family 35. The correlation in lots 3–5 comes out $+.319 \pm .082$, in lots 7–9, $+.539 \pm .069$ and in lots 10–14 $+.572 \pm .048$. For reasons which have been mentioned the latter two groups are much more satisfactory than the group of three miscellaneous lots, 3, 4 and 5. The average in lots 7–9 and 10–14 is $+.560 \pm .039$. The square of this coefficient, .314 indicates that over 30 per cent. of the variation in length of life is caused by heredity, neglecting such heredity as may be due to differences among families 2, 13, 32 and 39.

If we assume that some 10 per cent. of the variation is due to differences in condition, weight and age and over 30 per cent. to heredity, we still have over 50 per cent. of the variation due to unknown causes. From the nature of the case, however, a large amount of accidental variation is to be expected.

RELATIONS BETWEEN RESISTANCE AND OTHER CHARACTERISTICS

It is clear that family 35 is markedly more resistant to tuberculosis than the other inbred families, and that among these, family 2 is somewhat more resistant than families 13, 32 and probably 39. The question arises

whether these differences in resistance are related to any of the other characteristics in which these families differ.

We may dismiss color with a few words. Families 32 and 39 produce only animals of the primitive golden agouti color; families 2 and 13 produce only blacks, a color which differs from golden agouti by a single recessive factor; family 35 is composed of yellow agoutis. This color is recessive to golden agouti, depending on an allelomorph of albinism. We can not attribute the high resistance of family 35 to the yellow agouti color since the even more highly resistant crossbreds between family 35 and the other families are all of the golden agouti color also found in the susceptible families 32 and 39.

In all of the families there are varying amounts of white in a piebald pattern and of red or yellow in a tortoise-shell pattern. No influence can be attributed to the amount of white unless it is assumed that an intermediate condition is superior to either extreme. Family 39 has the least white, averaging less than 20 per cent.; families 13 and 32 have the most, averaging over 80 per cent.; while families 2 and 35 are intermediate with about 70 per cent. and 60 per cent. white, respectively. It may be added that the correlation between the amount of white in the coat and length of life in family 35, lots 10–14, was found to be virtually zero. Similarly, family 35 is intermediate between family 39 with least red and family 2 with most red in the colored parts of the coat.

The records in size, fertility and death rate among the inbred families and cross breeding experiments during the year 1919, in which all of these tested animals were born, is given in Table XVI. In this table, the weights and mortality records are corrected for the important effects of size of litter by calculating separately the averages in litters of 1, 2, 3 and 4 and finding an index in which these averages are weighted 1, 3, 3 and 1, respectively. This means practically that all of the records are reduced to the basis of an average size of litter of 2.50.

Because of the method of averaging, the figures are

not strictly comparable to those presented in Tables VII, VIII and IX for the animals actually tested. However, it is safe to conclude from a comparison of these tables that the animals tested were a fairly random selection from their respective families and experiments.

It may be noted in passing that the figures in Table

TABLE XVI

AVERAGES OF THE CHARACTERISTICS OF GUINEA PIGS BORN IN FIVE INBRED FAMILIES (2, 13, 32, 35 AND 39) AND CERTAIN CROSSBREEDING EXPERIMENTS IN 1919

CO is the first cross between inbred families. In *CA*, the sire is crossbred, dam inbred. *AC* is the reverse of *CA*. *B* is the control stock. The mortality and size characters are indices in which the averages for litters of 1, 2, 3 and 4 are weighted 1, 3, 3 and 1 respectively.

Family or Experiment	Number Young	Per Cent. Born Alive	Per Cent. Raised of Those Born Alive	Per Cent. Raised	Birth Weight (Grams)	Gain per Day to 33 Days (Grams)	Weight at 33 Days (Grams)	Size of Litter	Litters per Year	Young per Year	Young Raised per Year
2.....	331	80.3	88.9	71.4	76.9	3.40	189.1	2.43	3.82	9.30	6.46
13.....	259	87.1	85.2	74.2	88.4	4.76	245.4	2.85	3.79	10.79	6.67
32.....	146	90.3	78.8	71.2	82.2	4.13	218.6	2.25	3.48	7.81	5.30
35.....	258	80.2	84.7	67.9	88.3	4.57	239.1	2.37	3.68	8.72	5.64
39.....	72	93.7	80.7	75.6	77.9	3.79	203.1	2.40	2.97	7.13	5.15
Tot. Inbred.	1,066	84.0	85.1	71.5	82.8	4.12	218.9	2.47	3.63	8.97	5.96
CO.....	280	91.1	89.4	81.4	81.5	4.52	230.5	2.55	3.90	9.93	7.87
CA.....	302	85.8	94.5	81.1	84.3	4.53	233.8	2.72	4.24	11.53	9.20
AC.....	419	87.7	90.9	79.7	88.5	4.54	238.2	3.27	4.16	13.60	10.78
B.....	374	90.0	84.9	76.4	90.2	4.94	253.2	2.90	3.76	10.90	7.96

XVI give a good illustration of the statements made in regard to the effects of crossing, with the exception of a few records based on inadequate numbers.

From this table it will be seen that family 35, the most resistant to tuberculosis, held a rather low position among the inbred families in most other respects. It is actually the poorest in mortality among the young, and fourth in size of litter and first in nothing. There was thus no close relation between high resistance to tuberculosis and vigor in other respects in 1919.

However, the rank of a family in a single year is not always a safe indication of its true position genetically

in characters in which environmental conditions are of very much more importance than heredity. The rank of the families has been calculated for the periods 1906–1910, 1911–15 and 1916–19, making due corrections for the effects of size of litter. As previously stated, high correlations were found between the ranks in the first two periods in most respects among the 23 inbred families. Table XVII shows the ranks of the 5 surviv-

TABLE XVII

RANK OF FAMILIES IN RESISTANCE TO TUBERCULOSIS IN 1919, AND RANK IN OTHER CHARACTERISTICS DURING TWO PERIODS, 1911–15 AND 1916–19

Mortality and size characters are corrected for the influence of size of litter and (in 1916–19) for seasonal differences.

Family	Resistance to Tuberculosis	Per Cent. Born Alive	Per Cent. Raised of those Born Alive	Per Cent. Raised	Birth Weight	Gain to 33 Days	Weight at 33 Days	Adult Weight	Size of Litter	Litters per Year	Young per Year	Young Raised per Year
35....	1	3–3	3–1	3–1	2–2	2–2	2–2	2–2	1–2	2–2	1–1	1–1
2....	2	5–5	1–2	4–2	5–5	5–5	5–5	5–4	4–4	1–1	4–2	3–2
13....	3	2–4	2–3	2–4	1–1	1–1	1–1	1–1	2–1	3–4	2–3	2–3
32....	3	4–2	5–5	5–5	3–3	4–3	4–3	4–5	5–5	4–3	5–4	5–4
39....	3	1–1	4–4	1–3	4–4	3–4	3–4	3–3	3–3	5–5	3–5	4–5

ing families in the second and third period. The reality of the differences among these families is evident. Taking the following 6 characters—percentage born alive, percentage of these raised, birth weight, gain to 33 days, frequency and size of litter—the correlation between the ranks in the two periods is + .83.

The positions of the families agree in the main with those for the single year 1919. The position of family 35, however, is much better in most respects, a point which will be discussed later.

The difficulty of classifying the families in the order of general vigor is shown by this table. Families 2, 13 and 39 present curious combinations of high vigor in certain respects with weakness in others. It is true that there is perfect correlation between the ranks in size of litter and adult weight, but the order in which this places the

families, 13, 35, 39, 2 and 32, is far from being the order in frequency of litter or the mortality records. Taken as a whole, family 2 seems to have been rather the easiest family to keep going. Its regularity in producing litters and success in raising the young which are born alive are factors in this, but even more important seems to be another factor, probably correlated with that last named, the longevity of the animals after the matings are made, in which it far surpasses all of the others. Families 13 and 35 have also been relatively easy to maintain. This leaves families 32 and 39 as those most difficult to keep up to a desirable strength.

In spite of the better record of family 35 in the four years, 1916–19, as compared with the single year 1919, there is still no close relation between resistance to tuberculosis and vigor in other respects. In size, in both elements of fertility, and in the percentage of the young born alive, family 35 is still inferior to families which are distinctly more susceptible to tuberculosis. It may, however, be significant that family 35 led in the percentage raised of the young born alive and that family 2, second in resistance, is also second in this respect.

It may also be significant that while family 35 does not stand out in any particular element of vigor, unless in that last named, it stands relatively high in all, and so is the best family in the number of young produced per year by a mating, the product of frequency and size of litter, and is also the best family in the total percentage of the young raised, the product of the percentage born alive and the percentage of these raised. Moreover, family 2 is second in both respects as in resistance. These results suggest that while resistance to tuberculosis is not related to the most important factors which determine the various elements of vigor, it is a contributing factor to a sufficient extent to make the total efficiency of a resistant family higher than that of a susceptible family.

In this connection, the low standing of family 35 dur-

ing 1919 requires some consideration. The general condition of the stock was much better in 1919 than in any of the three preceding years. The principal cause of the high death rate, small and infrequent litters and slow growth during 1916, 1917 and 1918, especially during the first half of each year, was probably an insufficient supply of green feed during late winter and early spring. Symptoms, such as lameness, swollen and bleeding gums, were noted rather frequently. These were probably indicative of scurvy. A form of pneumonia was also rather common. Whether tuberculosis was present in the stock at this time is not certainly known. It is doubtful, as guinea pigs seldom take the disease unless directly inoculated. However, this may be, it seems probable that family 35 has a special ability to withstand exceptionally adverse conditions. It is not unlikely that this characteristic may be connected directly or indirectly with its resistance to tuberculosis. Under good conditions, on the other hand, there seems to be little if any relation between this form of vigor and apparent vigor in other respects.

Conclusions

There is little or no relation among guinea pigs between resistance to tuberculosis and sex. The present data indicate a possible superiority of the males, but one which is too slight to be certainly significant.

The data suggest a slightly greater susceptibility among the progeny of very young or very old females but this also is of doubtful significance.

Size of litter, birthweight and rate of gain up to weaning give virtually no indication of the probable length of life after inoculation.

The rate of gain preceding inoculation and the age and weight at that time all together determined less than 7 per cent. of the variation in length of life in a very heterogeneous lot of guinea pigs and less than 2 per cent. in a somewhat more homogeneous lot.

Marked differences in resistance were found among a number of inbred families of guinea pigs.

The high resistance of one of these families was transmitted by each sex to the offspring of each sex in crosses with other inbred families.

In crosses involving this most resistant family, the progeny were superior even to this family itself, indicating not only the dominance of resistance over susceptibility but possibly also the presence of complementary factors among the families.

In crosses among the more susceptible families, the progeny were little, if any, superior to the family of the better parent, indicating dominance but not complementary factors in this case.

Over 30 per cent. of the variation in length of life after inoculation among the crossbreds was determined by the amount of blood from the best inbred family. Allowing 10 per cent. of the variation as due to age, weight and condition, 50 to 60 per cent. remains as due to accidental and unknown causes.

The factors which determine the resistance of a family to tuberculosis are not closely related to the other elements of vigor, including rate of growth and adult weight, frequency and size of litter, the percentage of the young born alive and the percentage of these raised to weaning. There is some evidence that they are contributing factors to a sufficient extent to give the most resistant family a record above the average in each element of vigor and so give it the highest or nearly the highest total efficiency. Even this relation of resistance to other elements of vigor appears to be present only under exceptionally adverse conditions when it may play a direct part in determining the health of the stock.

The results in regard to resistance to tuberculosis are like those for other characteristics as regards the differentiation among families brought out by inbreeding, the improvement resulting from crosses between inbred families and the independence genetically from other elements of vigor.

GAMETIC AND OBSERVED RATIOS IN DROSOPHILA

DR. CALVIN B. BRIDGES

THE populations and families with which the geneticist deals are not the real objects of his investigation; for him, the distribution of characters is only an index of the preceding distribution of genes in gametes. But the whole course of embryonic development, with heavy mortality possible at every step, has intervened between the individuals that he classifies and the gametes from which they came. The observed classes correspond accurately to the original gametic series only in case this mortality is indiscriminate—that is, only if there is no differential viability.

In the breeding work with *Drosophila* there has been a continual effort to eliminate distortion in the ratios, which depends largely upon: (1) the extent of the mortality involved, this being characteristic in amount for each mutant type and character combination, (2) the suitability of the culture media and conditions, and (3) the competition when the number of developing individuals is in excess of the optimum number for the available food supply.

The problem of over-crowding (3) is simplest of solution, though over-crowding was the largest source of disturbance in most of the early work, as well as in some of the later. The remedy is, in the first place, to limit the number of eggs per culture to the output of a single female. No mass-cultures should be raised in experiments in which the ratios among the offspring are of importance. In the second place, as the larvæ grow larger and also increase in number with each day's output of eggs, the competition becomes intensified throughout the later stages of the culture. To meet this increasing demand, there must be fresh supplies of food, or enough food must be provided at the start so that even at the end there is sufficient for free development of all larvæ. In point of economy it is better to concentrate on a few cultures that are liberally supplied than to raise a greater

number that would mean optimum conditions for none and doubt concerning the reliability of all.

The main problem in connection with the environment (2) is to find a kind of food that will allow full development of even very weak classes. It is in this field that the greatest changes in method have been made. For some years—from 1910 to 1916—some modification of the fermented-banana method of preparing food was followed. Ripe sound bananas were peeled, and the pulp left for about 24 hours in a liquid containing yeast. This liquid was usually the fermented juice from the previous lot of bananas. About 25 grams of this fermented banana was put in the bottom of a culture bottle and covered with absorbent paper.

It was suspected that the real food of the larvæ was not primarily the banana but was rather the yeast cells and perhaps also the bacteria, the banana being mainly the culture medium for the yeast. This has been established by the work of Northrop,[1] of Loeb and Northrop,[2] and of Baumberger.[3] In July, 1916, in consultation with Northrop, I started experiments with a view to using as a culture medium standardized solutions, instead of banana. The solution was absorbed and held in a cake in the bottom of the culture bottle by shredded paper toweling, which offered extensive surface for the growth of the yeast. This method was unsuccessful; the flies laid few eggs and these were often overgrown by the yeast and killed. Esters and other chemicals with fruit odors did not lead to greater egg production. Perhaps better success with culture solutions would be obtained in supplementing and modifying banana methods.

The banana method was modified with a view to discouraging the growth of moulds and putrefactive bacteria by mild antiseptics or correctives, such as benzoate, thymol, formaldehyde, alcohol, powdered marble for neutralization of excessive acidity, etc. Good results were obtained with alcohol, where several extensive sets of comparative tests seemed to show that about 1.5 per cent. of alcohol in the food was desirable. The most successful alcohol method was roughly as follows: The pulp of sound ripe bananas was weighed and put with an equal number of c.c. of 3 per cent. alcohol in a shallow, covered dish. No yeast was added, since enough wild yeast was usually present. The food

[1] *Jour. Biol. Chem.*, 1917, pp. 181–187.
[2] *Jour. Biol. Chem.*, 1916, pp. 309–312.
[3] *Jour. Exp. Zool.*, 1919, pp. 1–28.

was at its best when it had fermented for about 24 hours. The optimum amount of drained banana was found to be about 25 grams per bottle. This was put upon the bottom of the culture bottle and one gram of paper-toweling strips (about 5. ×.7 cm.) was matted down on the top. Pint culture bottles gave a greater output per pair than halfpints. The alcohol method was used more successfully than the old method during the fall and winter of 1916.

In the spring of 1917 considerable work was done in testing out various media containing starch, sugar, peptone and salts. This method gave good results except that trouble with moulds was greatly increased.

In the autumn of 1916, Dr. R. W. Glaser told me of certain culture-media experiments that Mr. Baumberger and he were carrying out with banana infusions and agar.[4] Dr. Glaser later sent me directions for preparing these media and also some prepared tubes. My tests of the method showed that the amount of food was inadequate for general use, although sufficient for the small number of flies that they wished. I increased the concentration of the media by the addition of sugar, banana flour, etc., but principally by grinding up and using all the pulp of the bananas, instead of using simply the strained juice. A comparison of fresh banana with banana that had been fermented before incorporation showed that the fresh banana was superior. Likewise fresh banana was superior to banana raised to the boiling point at any stage of preparation. It was found that yeast should not be distributed throughout the media. Experiments showed that it is advisable to have a very light seeding of yeast confined to the surface of the solidified media. Also it is well to keep the yeast from the margin as much as possible, since fermentation at the sides and beneath the cake makes the cake break loose and rise. The amount of agar was found to be adequate at 1 per cent.

It was some months before this method was improved so far that it gave better results than those given by the old method or the alcohol method. In the spring of 1917 it was worked out well enough so that it was substituted for the old method in my regular work. By the winter of 1917 it had become quite

[4] *Science*, 1917, pp. 21–22.

generally adopted in the laboratory. Several points have been improved since, so that the procedure at present is as follows:

1. Use bananas that are thoroughly ripe or over-ripe.
2. Peel the bananas and weigh the pulp (100 grams of pulp provides for about four culture bottles).
3. Weigh agar-agar, 2 per cent. of amount of banana.
4. Measure as many c.c. of water as there are grams of banana.
5. Add agar-agar to water and heat until the agar has dissolved. (Complete solution is hastened by the addition of a small amount of fresh water soon after the boiling point has been reached.)
6. While the agar is heating, press the banana through a potato masher or a coarse sieve, and place in readiness the bottles (which should have been previously washed and also preferably steam sterilized). Get ready yeast (Magic Yeast ground up) and paper (absorbent paper, paper toweling cut into 4-fold squares $3'' \times 2''$) and cotton (stoppers may be reused, but should be dry sterilized by enclosing over formalin. Cotton stoppers are better if made rather tight and covered with very soft cheese-cloth).
7. Stir banana into hot agar solution. Mix thoroughly. Mixture should not be heated any longer.
8. With ladle and funnel pour about 50 c.c. of the media into each half-pint or pint milk bottle. (The media should be at least $\frac{3}{4}$ in. thick to stick well.)
9. Sprinkle top lightly with dry yeast.
10. Put in contact with media a 4-fold square of absorbent paper.
11. Stopper with cotton.
12. Use same day. Best to use as soon as cool. Not good after two days.

Flies can be mated in vials and then transferred to the culture bottles at the end of the day. A little food may be kept going by the alcohol method for use in vials, for covering over mould patches in culture bottles, and for refeeding stock cultures.

The distortions in the ratios that arise from mortality characteristic of given mutants and combinations (1) can not be eliminated by direct methods. Fortunately, a large proportion

of the mutants are little if at all below normal in degree of viability; that is, when such mutants are compared with the wild type under identical conditions, the observed ratios show little or no deviations from expectation beyond those due to random sampling. As an example may be mentioned white-ocelli, which is known to have maintained itself with practically undiminished frequency through 175 generations of competition, under unfavorable conditions of culture,[5] with the wild-type. In the main, the mutant races that show normal viability are those whose somatic effects are "slight". Thus, white-ocelli affects the color of the tiny group of three ocelli on the top of the head. The character, though involving so small an area, is perfectly sharp and definite, and under proper conditions of illumination and magnification is fairly easy to classify. The same is true of many other "slight" mutations, such, for example, as speck, cross-veinless, and hairy, which are among the most valuable *Drosophila* mutations. On the other hand, mutants that involve more extensive or manifold changes, such as club, notch, rudimentary, and delta, are also among those poorest in viability. Some of these changes in themselves interfere with the success of the individuals possessing them: flies with "spread" wings or "dachs" legs are liable to become caught in the culture media and die. These changes are also sometimes obviously accompanied by serious internal derangements. In the case of streak, for example, it can be seen that the internal muscles of the thorax are largely replaced by bubbles and blood sinuses. The correlation between inviability and the extent of the visible change is high, but is lessened by the cases in which the internal accompanying changes are of slight disadvantage. Thus, the mutant "pads" resembles "club" very much, and appears to be a greater change in the same direction, but is nevertheless far freer from inviability. Conversely, certain mutants that are usually lethal occasionally do produce offspring, which are then not as strikingly different from the wild-type as some other mutants that have good viability. Lethal-10 very occasionally survives, and the individuals are scarcely to be distinguished from dwarfs of a certain mutant race (dwarfoid) that is little inferior to the wild-type in viability.

[5] *Biol. Bull.*, 1920, pp. 231–236.

The connection between observed character-change and inviability is even more indirect than suggested above. In the *Drosophila* work it is not the comparative viability of adults possessing given character differences that is of the most importance. Even though many of the characters are of such a nature that their possessors would be under a serious handicap in competition, in relatively few cases does this fact lead to alterations in the observed ratios, since the classifications are made usually soon after the flies hatch, *i.e.*, every 24 to 48 hours. It is true that certain mutant forms such as "divergent" and "gull" and "bifid" wings, also "dachs" and "reduplicated" legs tend to become entangled in the culture media and drowned immediately after emergence, so that in these cases the observed ratios are somewhat different from the hatching ratios. There are also a few mutants—mostly semi-lethals—in which the adult is unable to live very long even under the most favorable conditions. Among these may be mentioned "lemon," "apterous," and especially "decrepit." The "decrepit" flies die a few hours after hatching in spite of all care in helping them emerge from the pupa case, in keeping them in quarters not too dry or wet, and in suplying them with suitable food. It would seem that the death of such flies as are obviously weak on hatching is to be referred to difficulties encountered in the pupa stage.

Even inviability arising in the pupal stage, like that in the adult stage, is less general and significant than that in the larval stage. Most of the inviability that affects the ratios of adults is to be referred to differences acting in the larval stage, as is evident from comparative studies of the results of pair and mass cultures and of changes in culture methods that affect only the larval period. The difference between mass and pair cultures is essentially a difference in the number of larvæ that are in competition, the food conditions and the character of the larvæ being at first identical in the compared cultures. It is found that the distortion to the ratios among the adults is roughly proportional to the number of larvæ in competition. How extreme such competition may be is evident from the fact that a point is soon reached after which further increase in the number of mothers brings no increase in the number of progeny and may even result in a decrease. So predominant is the larval stage in its influence upon viability that the chief field of

improvement of culture conditions has been that of the character and methods of use of the food for the larvæ. There are specific viability differences among the larvæ of the different mutant types and combinations. Such viability differences must depend upon differences in the characters of the larvæ, and these, because of the intervening metamorphosis, have little direct relation to the characters of the adult, but are products of the action of the same mutant gene. The high correlation observed between extensive change in adult characters and high degree of inviability must, then, mean that such genes generally cause changes which interfere directly with the success of the larvæ.

Three larval characters are known—the tumor responsible for the death of lethal-7 larvæ, the much shortened larvæ of the mutation "chubby," and a marking on the posterior end, viz., "barette." It is supposed that a high proportion of the larval characters that lead to inviability are differences in internal structures, but some of these might be detected. However, no systematic search for larval characters has been made even in the case of the inviable mutants where such differences are probably present.

As we have stated, the distortion in ratios that arises from inviability, especially inviability originating in the larval stage, can be very materially reduced by improvements in culture media and in methods. Many poorly viable mutants can be made quite generally usable, as, for example, dachs. But when a mutant such as dachs is to be used in linkage determinations, the experiments should be so planned as not to include more than one of these characters. The presence of a single poorly viable character in an experiment does not prevent the calculation of correct crossover values. The complementary classes that do not include the inviable character should be in the same proportions as in the gametic series. Even in cases of mutants completely lethal, the linkage relations of the lethal gene can be calculated accurately from the ratios shown by the other characters of the cross. The classes that include the inviable character are often also usable, but with less certainty that the values are correct. Such values are correct when the presence of the mutant decreases by the same percentage the size of every class in which it occurs. The fewer the mutants involved in an experiment, the greater the likelihood that this

result will follow. Unexpected irregularities in ratios may arise where many mutant characters are distributed in different combinations. These peculiarities of inviability are probably comparable to "specific" and "disproportionate" modifications in eye-color, etc.[6]

If more than one poorly viable mutant is present in a linkage experiment, there is distortion in the ratios due to linkage, and such experiments are either entirely worthless, or are only to be regarded as rough indicators of the real relations. As we saw, if only one inviable mutant is present in a cross, one of each of the pairs of complementary classes remains undisturbed; and correct values can be calculated from them. The presence of a second inviable mutant leaves undisturbed only that class in which neither mutant occurs. The calculation of crossover values under this circumstance is somewhat comparable to solving for two unknowns with a single equation. Solutions can be obtained only by assuming some relationship between the two disturbances. Thus, we may assume that the disturbances are independent; that is, that there is no specific interaction of the kind mentioned above, and the class in which both mutants occur is accordingly of the size that would be expected from the amount of the disturbance present in those classes in which each occurs by itself. On this basis, the crossover values are calculated from the square root of the product of the two complementary non-crossovers, and likewise of the crossover classes, instead of from the sums of such complementary classes.[7] The assumption of independence would be approximately correct in perhaps a majority of crosses in which only two or a few loci are involved. If the disturbances are related—if they tend to neutralize or to exaggerate each other—a correction can still be made by raising an equal number of individuals in the complementary cross. In the two complementary back-crosses, a $\times$ b (repulsion) and a b $\times$ wild-type (coupling), the character combinations that are non-crossover classes in the repulsion experiment are crossovers in the coupling experiment, and vice versa. If the presence of a particular class has given a crossover value too high in the one cross, then it will give a value correspond-

[6] *Jour. Exp. Zool.*, 1919, p. 374.

[7] This geometric mean method was proposed by Muller, who gave an excellent discussion of the difficulties involved in differential viability. (AM. NAT., Vol. L, p. 351 ff.)

ingly too low in the complementary cross, and the mean value will be correct.[8]

Since the effects of inviability are likely to be more pronounced, even disproportionately so, as the number of mutant characters present simultaneously is increased, it is advisable to plan any linkage experiment in which several characters are to be involved, in such a way that the characters are distributed as evenly as possible. The type of back-cross that gives the evenest possible distribution, as well as the smallest proportion of individuals in which the higher combinations occur, is that in which half the mutants have entered the cross from one parent and the other half from the other parent, and in which the mutants are *"alternated"* as regards their positions along the chromosome. Thus, for example, let us consider a back-cross in the third chromosome in which the seven mutants to be used are: roughoid at 0.0; hairy at 25.8; scarlet at 35.1; dichæte at 38.5; pink at 44.6; spineless at 54.2; and ebony-2 at 66.9. The two parents should be roughoid scarlet pink ebony-2, and hairly dichæte spineless; and the formula for the F_1 multiple heterozygote would be:

$$\frac{\text{ru} \qquad \text{st} \qquad \text{p} \qquad \text{e}^2}{\text{h} \qquad \text{D} \qquad \text{ss}}.$$

The production of an individual possessing all seven characters would require an hexuple crossover, which almost certainly would not occur.[9]

A method that overcomes inviability effects to the greatest extent where many mutants are involved, but which unfortunately requires too great labor for general use, was devised by Muller.[10] F_1 females heterozygous for any number of mutants are crossed, not to the multiple recessive as in the ordinary back-cross, but to the wild-type. Except for the dominant mutants, none of the characters involved appears in the resulting individuals, and hence do not exert injurious effects. These indi-

[8] This "balancing of the inviability" has been discussed at greater length by Bridges, *Jour. Exper. Zool.*, 1915, p. 3 ff.; *Jour. Exper. Zool.*, 1920, pp. 281, 288; by Morgan and Bridges, Carnegie Pub. No. 237, p. 19, 43; and by Muller, AM. NAT., 1916, p. 353.

[9] See Bridges, *Jour. Exper. Zool.*, 1920, p. 295, for discussion and other examples of the "alternated back-cross."

[10] AM. NAT., 1916, p. 354 ff.

viduals have then to be tested singly to determine what recessive characters they carry and hence to what crossover category they belong.

Thus, by improvements (1) in the type of experiment planned, (2) in the culture media and methods used, and (3) in the method of calculation, disturbances in the ratios can usually be held within negligible amounts.

To these indirect methods of obtaining accurate values is to be added one still more important—namely, the discovery of new mutants in which viability is practically normal, and which can be substituted for mutants less satisfactory in that regard. Many of the loci are represented by several mutant allelomorphs, which often are different in viability as in other characteristics. Thus, of the eight cut allelomorphs, or appearances, cut-6 is distinctly the most nearly normal in viability. Likewise, of the five or six allelomorphs of the truncate locus, "dumpty" is the most satisfactory.

In most of the more complex linkage problems, especially those involving linkage-variations or coincidence, the behavior of particular regions of a chromosome is being examined, and the particular loci utilized are only indices of the behavior. What is most essential, therefore, is that there be workable mutant loci distributed rather evenly over all regions of the chromosomes. As the number of mutants in a particular region increases, there is a greater range of choice and greater probability that one or more of the mutants of that region will have normal viability. Thus, bifid (at 7.3) was long the only workable mutant at a favorable distance from the left end of the X-chromosomes. More recently ruby (at 7.5) because of its better viability has displaced bifid from general use, and this in spite of the fact that ruby interferes with the classification of several other eye-colors (especially prune and garnet) while bifid is workable with nearly all other mutants.

In regions less well represented in numbers of mutant loci, a mutant with excellent characteristics may be used rather than one whose position is more favorable but whose other characteristics are poorer. Thus, "humpty" is very favorably located in the second chromosome in the middle of the long region from curved (73.5) to plexus (98.5), but its viability is so poor that in most experiments it is better to leave this section unfollowed than to introduce humpty. There are at present few regions

that are not satisfactorily represented. By far the longest of these in the 25.8 unit interval from roughoid to sepia in the left end-region of the third chromosome. Because of uncertainties in classification, it is not ordinarily possible to use more than two eye-color mutants together in an experiment, and such masking of one character by others affecting the same organ leads to a continued search for combinations of characters that can be handled simultaneously. In general, the slight mutant characters mask each other less than do extreme ones, and these are, usually the least inviable. There is a continual improvement of the working material by the substitution of better mutants.

A PROBABLE EXPLANATION OF POLYEMBRYONY IN THE ARMADILLO

PROFESSOR CHARLES R. STOCKARD

CORNELL UNIVERSITY MEDICAL COLLEGE, NEW YORK CITY

BY arresting the development of the fish's egg during early stages double individuals and twins are frequently induced. The interruption or arrest makes it possible for more than one potential growth point along the germ-ring to give rise to an embryonic shield. In other words, accessory invaginations or blastopore formations occur as the initial structural step in doubleness. The interruption in the development of the fish embryo must be introduced during the cleavage stages and before gastrulation in order to produce such phenomena. Among hundreds of eggs arrested during later developmental stages no double monsters or twins ever occurred. A complete account of these experiments is soon to be published but for our present purpose two facts are important: First, accessory embryo formations result from arrests in the developmental process; and second, the arrest must occur before gastrulation has taken place.

In the light of these experiments it has seemed possible to interpret somewhat more clearly than has formerly been done the remarkable phenomenon of multiple embryo formation in the armadillo.

On examining the uterus in two pregnant specimens of a South American armadillo von Jhering, in 1885, discovered that each contained eight fetuses enclosed within a single chorion. He correctly concluded that all of the fetuses in each mother had been derived from a single egg by some process of division into separate embryonic rudiments. After this valuable discovery and interpretation, the study of the armadillo's development lapsed and nothing of importance was added for almost twenty-five years. Two series of investigations were then begun simultaneously one on the Texas armadillo by Newman and Patterson,[1] and the other on the South American species by Fer-

[1] H. H. Newman and J. T. Patterson, *Jour. Morph.*, Vol. 21, p. 359, 1910.

nandez.[2] The growth and expansion of these twin studies has brought our understanding of the phenomena of polyembryony in the armadillo to a considerable state of maturity.

These authors readily agreed that in most species of armadillo the individual members of a litter, usually four in the Texas species and eight in the common South American form, are all derived from a single egg. It required considerable effort, however, to obtain the material that would furnish the morphological stages of the process by which the polyembryonic development was accomplished. We are finally indebted to Patterson,[3] for the very thorough and satisfactory manner in which he has collected and studied the early embryonic conditions; and particularly for having shown the first stages of the budding process through which the single blastocyst gives rise to four distinct embryonic areas, each exhibiting a typical primitive streak region.

In connection with the fish experiment it now becomes important to ascertain exactly what degree of development has been attained by the armadillo blastocyst at the time the budding process begins. And since, according to my interpretation, these buds should arise at the time of gastrulation or blastopore formation, it becomes necessary to consider very briefly the germ-layers and gastrulation in mammals. The decidedly precocious and highly modified method of forming the primary germ-layers in the mammalian blastocyst is not strictly comparable to gastrulation or the method of germ-layer formation found among the other vertebrates. On the other hand, the embryonic line or primitive streak of the mammalian egg is exactly comparable to the blastopore and head process formation in the simpler forms.

The blastocyst of the armadillo has already, by a process of cell migration and delamination, separated off the primary entoderm from the ectoderm and further modified these layers before the budding which forms the embryonic primordia has begun. The primordia are first formed by a thickening of the ectodermal layer of the blastocyst. The primary entoderm then invaginates into the primordia to form the secondary entoderm of the gut. The precocious cell migration and splitting into layers in the mammal's egg is associated with the early implanta-

[2] M. Fernandez, *Morph. Jahrb.*, Bd. 39, p. 302, 1909.

[3] J. T. Patterson, *Jour. Morph.*, Vol. 24, p. 559, 1913.

tion of the embryo upon the uterine-wall of the mother, and the later primitive streak formation may be interpreted as related to the actual gastrulation or blastopore formation away from which the line of the embryo always develops.

Whether the validity of the above briefly outlined interpretation of the germ-layer formation is admitted or not, we have in the armadillo a process of budding taking place from the blastoderm and associated with accessory or extra blastopore formation in much the same way as are the accessory embryos along the germ-ring in the egg of the bony-fish. These buds also accord with Kopsch's description of a double gastrular condition with two blastopores in a blastoderm of Lacerta agilis, from which he concluded that twin formation as well as anterior duplication arises from a double Einstülpungen. And further, Assheton has described a similar condition in a blastodermic vesicle of the sheep. He, however, imagined the condition to have been due to a splitting during the morula stage.

The double primitive streaks in the hen's egg and other forms all lend themselves to strengthen the interpretation that double embryo formation first asserts itself by a double gastrulation or blastopore formation, which is initially a process of double instead of single bud formation. Patterson's description of the origin of the quadruplet buds in the Texas armadillo furnishes the most striking case in the study of these conditions. And we may conclude that the budding or accessory embryo formation in the egg of the armadillo is exactly the same developmental process as that which gives rise to twins and double individuals in other vertebrate eggs.

However, the very important question yet remains to be answered. Why does this accessory bud formation occur so constantly in the Texas armadillo in contrast to the single embryo formation of mammalian eggs in general? Patterson failed to answer this question, but he supplied some very significant data which Newman,[4] has appreciated as being intimately connected with the occurrence of polyembryony.

In connection with the collection of material Patterson[3] discovered a "period of quiescence" of the embryonic blastocyst. Regarding this he states:

> The fact was first made apparent in 1911, when, after I had started collecting two weeks earlier than in the preceding year, I failed to

[4] H. H. Newman, "The Biology of Twins," Univ. of Chicago Press, 1917.

obtain the cleavage stages, although judging from the condition of development in the vesicles collected in previous years, one would naturally expect to find these early stages during the period of my first collection in 1911.

The following year he began collecting still two weeks earlier and again had a similar experience.

Practically all of these vesicles lie free within the uterine cavity, either in the horizontal groove or in the region of the attachment zone (placental area).

It is evident from these data that the embryonic vesicle remains for some time lying free within the uterine cavity. Just how long this period lasts, I am unable to state; for practically every old female taken at the earliest date (October 15) at which I have collected, possesses a free blastocyst. . . . Taking all the facts into consideration, I estimate the "period of quiescence" to last about three weeks; that is, from about the middle of October to the third or fourth of November.

In a study of sections no mitotic divisions were found to occur in the blastocysts during the "quiescent period."

The only point of interest cited by Patterson in connection with this peculiar phenomenon of interruption in development, was the fact that in no other mammal, except the deer, had such a condition been found. Bischoff had long ago, 1854, reported a "period of quiescence" lasting for some weeks during a so-called morula stage of the deer embryo.

Newman[4] has recognized the importance of Patterson's discovery of a "quiescent period" during the early development of the armadillo, and states in a discussion of twin formation that this "period of quiescence" probably, "holds the clue to the physiological explanation of polyembryony." In this position Newman is, in my opinion, largely right, but this is as far as the data led him, and he finally remarks:

The problem is to locate the factors responsible for the slowing down of the developmental rhythm. Whatever these factors may be, and we have no definite knowledge of them, the result of retardation is polyembryony.

Newman thus fails to appreciate the second point in Patterson's discovery, and that is that the blastocysts always lie free in the uterus during the "period of quiescence." This fact enables us to go one step further since the lack of attachment and, therefore, lack of oxygen supply are very probably "the factors responsible for the slowing down of the developmental rhythm."

The armadillo egg like that of most mammals undergoes its early development in the fallopian tube and is, therefore, capable of reaching the blastocyst stage on its initial oxygen supply. After this time, however, it must become attached to the uterine wall for a further source of oxygen. For some reason in the armadillo the reaction between the blastocyst and the uterine wall is postponed, and the blastocyst is incapable of further developmental progress until this reaction is established and the necessary supply of oxygen becomes available. In exactly the same way the development of the blastoderm in the fish's egg is experimentally retarded or stopped by reducing the available oxygen supply and is again made to resume its development by supplying oxygen. In the case of the fish egg, the supply of ordinary nutriment is certainly not involved, and reactions similar to those of the armadillo egg are only obtained as responses to changes in temperature and rate of oxidation.

In the armadillo egg I also do not believe the retardation is of the nature of a starvation phenomenon, since we see nothing of the kind in other forms. Temperature changes are ruled out, since the temperature of the uterus is more or less constant. The absence of oxygen necessary for the energetic process of cell division, is, therefore, in all probability the arresting cause, and the retardation results in polyembryony.

Thus Patterson has found the developmental interruption to exist, and he has also shown the blastocyst to be disconnected from the uterine wall and its necessary oxygen supply during this time. However, he has furnished no data bearing on the reason for the delay in uterine reaction and the consequent failure of immediate implantation of the blastocyst such as normally occurs in other mammals. However, from what is known of the dependence of uterine reactions on conditions in the ovary (Leo Loeb,[5] Stockard and Papanicolaou[6] and others) it may very probably be that some peculiarity in corpora lutea formation is primarily responsible for the entire series of reactions leading to polyembryony in the armadillo.

The consideration of the armadillo egg up to this point has taken account only of the external factors influencing its mode of development. It must now be remembered as a fact of serious

[5] Leo Loeb, *Jour. Morph.*, Vol. 22, 1911.

[6] C. R. Stockard and G. N. Papanicolaou, *Am. Jour. of Anat.*, Vol. 22, 1917.

importance that the production of quadruplets from the single egg of the Texas armadillo is an almost constant occurrence, while the experimental attempts to produce twins and double individuals in fish eggs and other forms have given at best only small percentages of such individuals among the large groups of eggs treated. It is also a fact that all eggs do not furnish equally favorable material for artificial twin production. The eggs of the trout seem unquestionably more disposed to give rise to twin formations than do the eggs of Fundulus. Thus some eggs would seem to have a hereditary or truly innate predisposition towards polyembryonic formations. There is much reason to believe that aside from the external factors discussed, the armadillo egg itself is highly disposed toward the formation of accessory embryonic buds.

There is the possibility, of course, that this natural experiment with the armadillo egg has become so exactly regulated as to influence the developmental processes precisely the same way each time, yet this is highly improbable. The armadillo egg is not a case of simple twin growths from the blastoderm, but as Patterson finds, there are primarily two buds, and then very promptly two secondary ones arise making the four and after this the budding process ceases. In the South American species, however, it would appear as though a tertiary budding occurred giving the usual eight embryos; and in rare cases still another budding occurs from a few of the existing buds giving a total of as many as twelve. It would certainly seem as though the blastoderm in these species passes through a stage of agametic reproduction or budding of a nature unknown among other higher vertebrates. But the possibility for such expression might only exist on account of the delay in implantation of the blastocyst and consequent shortage of the oxygen supply necessary for the rapid formation and growth of the single embryo.

It is important to keep in mind that there are species of the armadillo which produce only a single offspring from one egg. It is not known whether their embryos have a "period of quiescence" but if they have, the period either occurs at a different developmental stage or the eggs do not possess the inherent budding tendency of the other species.

We have further to acknowledge the fact that although the egg of the deer has a "period of quiescence" during its development it does not give rise with any degree of frequency to twin indi-

viduals. In the first place it is entirely uncertain from the scanty accounts as to what time in development the quiescent period occurs. Assuming that such a period does exist, it might occur at some indifferent stage when no peculiar result would be expected, for example after gastrulation, as it does in the bird with no subsequent effect. In the light of the experimental production of double individuals it is readily understood that even though the egg of the deer is interrupted in its development at an early stage, it might still be capable, on resuming development, of giving a normal single embryo. A study of the experimental production of twin and double individuals among fish leads one to be surprised at the case of the armadillo, and to expect the reaction found in the deer. The constant interruption occurring in the development of the birds and other animals at indifferent developmental moments with no subsequent ill effects, renders commonplace the fact that the deer successfully withstands an interruption during its development without noticeable modifications in structural response. A full consideration of the different results following interruptions at critical and indifferent developmental moments will be published in a forthcoming number of the *American Journal of Anatomy*.

In conclusion we may summarize the cases as follows: The development of the armadillo is interrupted on account of a failure to become promptly implanted on the uterus and a consequent exhaustion of the available oxygen supply. The interruption occurs at a critical period just preceding the primitive streak and embryonic line formation. This egg appears to have a decided tendency under conditions of arrest to form accessory embryonic buds. As a result of the interaction of these external and internal forces polyembryony is produced.

In the case of the deer only one probable fact is known, and that is that a "period of quiescence" occurs. It is uncertain at what stage the arrest takes place but it is probably due as in the armadillo to a delayed implantation of the blastocyst. Either on account of the stage of arrest, or a lack of tendency to form accessory embryo-buds a typically single individual arises from this egg. The external factors may be the same as in the case of the armadillo, but they interact with different internal factors or different developmental moments to give a very different result.

NOTES AND LITERATURE

North American Early Tertiary Bryozoa. By FERDINAND CANU AND RAY S. BASSLER. Smithsonian Institution. United States National Museum. Bulletin 106. 1920, 879 pages, 279 text figures and 162 plates.

Students of both fossil and recent bryozoa will greet with interest and pleasure this monumental work long anticipated and recently issued, for while treating primarily of fossil bryozoa this monograph contains much of interest to students of living forms. This work appears in two volumes, one containing the text and text figures, the other consisting of photographic plates alone. A cursory inspection reveals the fact that these volumes possess the excellence of copious illustration, a most satisfactory virtue in the eye of those who will use them. The text figures are abundant, the number as stated above (279) by no means giving a true idea of the actual number, since each figure consists of from two to ten or more illustrations, representing portions or organs of the species under discussion, and often besides figures of nearly related species for comparison. From this point of view the number given should be multiplied many times, and by actual count the first fifteen text figures contain more than one hundred separate drawings or preparations. These are all either original with the authors or are taken from the illustrations of other bryozoologists. Each photographic plate likewise contains from twelve to twenty-five separate photographs. These are distinguished by a remarkable clearness and definiteness of outline, even of minute details, revealing an unusually skilful management of light and shade and producing an excellent and expert piece of work which will not fail to call forth the gratitude as well as the admiration of their fellow workers.

Over 700 species belonging chiefly to the two orders, Cyclostomata and Cheilostomata, are treated in this monograph. In the seventy or more pages of introduction the authors present many topics of interest involving new points of view which will doubtless stimulate further research. Of these topics but three will be touched upon.

1. It is gratifying to find clear definition and illustration of

the terms of the more recent nomenclature used in description and classification. The study of the bryozoa has developed to such a degree in recent years and so many new terms have been introduced which are found only in the scattered writings of numerous authors, that a new compilation and exposition similar to the classic work of Hincks in his "British Marine Polyzoa" would be useful. As far as it was compatible with the limits of this treatise such a compilation and exposition have been accomplished here, and both the beginner in the study of the bryozoa and the advanced worker will find great assistance in the discriminating use of the newer anatomical terms.

2. Of the general functions of the bryozoa, of the Cheilostomata especially, the discussion of the hydrostatic function is perhaps the most interesting because it presents the newest and latest views on this puzzling subject. The extrusion and retraction of the polypide, the action of the operculum and of the zoœcial muscles in these activities, and the relation of these to the ingress and egress of water was long a puzzle. Jullien in 1888 first discovered the so-called compensating sack or compensatrix under the dorsal surface of the zoœcium. Since then scattered studies have been made on this organ which was soon found to be present in many species. The present authors have continued this study and following Levinsen (1909) have made the presence or absence of a compensatrix the basis of division of the Cheilostomata into two sub-orders Anasca, without such a compensating sack; Ascophora, possessing such a sack.

In addition to the zoœcial hydrostatic system discovered by Jullien, the senior author, in 1915 discovered a zoarial hydrostatic system in the Anasca. This investigator found that the space under the ectocyst, in certain species lacking a compensatrix, was continuous from zoœcium to zoœcium throughout the colony. Into this space water is introduced or expelled thus compensating for the egress and ingress of the polypides. By such means minute creeping zoaria as the Lunulites, *e.g.*, are enabled to maintain themselves on the algæ on which live.

3. In line with their insistance on the value of function it is not surprising that these authors classify the Tertiary bryozoa on a physiologic rather than a morphologic basis as is the method followed by the older investigators. Believing as they do that (p. 70) "In the bryozoa, as in other living beings, the form is only the result of functions; therefore in the study of

morphological variations of the organs, we now substitute that of the physiologic functions. Our studies are therefore always directed toward the discovery of functions which modify the skeletal form." With this frank statement of the primacy of the Lamarckian principle, family, genus and species are thus briefly described:

The family is characterized by the larval form.

One genus differs from another in possessing a different function. The three essential functions of all bryozoa are:

1. Passage of egg and escape of larvæ (= rapport of operculum and ovicell).
2. Hydrostatic system and extrusion of the polypide (= form of the aperture and rapport of operculum with compensatrix).
3. Calcification and chitinization (= nature of the skeletal part and of the frontal considered as deposits of the endocyst).

Specific characters include all morphological variations and all of the characters of adaption.

Whatever philosophic views one may hold in regard to the relation between form and function, it is apparent that the characters chosen for family, genus and species present a uniform, logical system and constitute a good workable plan or hypothesis which it seems more than worth while for all workers in this field to attempt to apply. It must be remembered, however, that while in the class bryozoa, larval characters may afford valid data indicating relationship such characters have failed to afford satisfactory data of relationship among some other class of animals. Granting the validity of the assumption, however, the research necessary to establish this statement can be conducted only on living species. This, then, is a matter of immediate and pressing interest. The older bryozoologists considered the operculum a family character. In the present system the operculum becomes a generic character, changes in it being induced by changes in the essential zoœcial functions 1 and 2, that is, depending in part upon the relation existing between the operculum and ovicell, and in part upon the relation existing between operculum and compensatrix together with the form of the aperture. Here again, although much investigation has been conducted to verify these generic principles among the fossil Tertiary species, work on living forms should be undertaken to

discover how far recent species actually conform to the plan proposed.

Identification and description of the species of this collection constitute the main portion of this monograph. A superficial examination of the text reveals the fact that in order to apply these principles to present day bryozoa, momentous and wide spread changes will be necessary. Not only will genera and families be broken up, but many heretofore considered widely separate will be regarded as closely related and vice versa. While one is startled by the number and significance of the changes involved, yet the present morphologic method is so unsatisfactory that this attempt to apply a unitary principle which promises so to simplify classification and to lift it out of chaos, should be heartily welcome. Too high praise cannot be accorded the authors of this monograph for the excellence of this work so full of new and stimulating ideas.

ALICE ROBERTSON

SEATTLE, WASHINGTON

SHORTER ARTICLES AND DISCUSSION

THE EFFECT OF YEAST ON THE UTILIZATION OF FOOD BY WHITE MICE.[1]

In the present paper the question of the effect of the so-called vitamines on basal metabolism is considered and a procedure is indicated whereby it is believed more conclusive data may be obtained on the question. Preliminary experiments are described illustrating the method.

Several years ago Hopkins[2] in a carefully carried out experiment investigated the effect of a small addendum of milk to a diet of purified food stuffs. In brief his method was to feed in pairs two sets of young rats of the same origin, weight, etc., on a basal vitamine-free food and to one set give a small addendum of milk and determine the food intake and growth increment. By comparing the energy consumption and growth increments of the two sets of animals and by comparing these factors on the same set of animals after reversing the diets he was able to show "that the small milk addendum reduced the food consumption for a given weight increment to one half or less." In other words the vitamine increased greatly the animal's power to utilize its food in the production of growth. And as he showed that this was not due to difference in absorption from the intestines, the vitamine must have an effect on some factor involved in basal metabolism.

A number of the criticisms that could be made of Hopkins' method have been discussed frankly by him and there is left little or no doubt concerning his conclusions. They have, however, such an important bearing on problems in nutrition that it seems very desirable that the question be investigated from all angles[3] and particularly with other animals,[4] and with vitamines

[1] Contribution from the Department of Pharmacology, Harvard Medical School.

[2] *J. Physiol.*, 1912, 44, 425; see also *Biochem, J.*, 1913, 7, 97.

[3] Recently Eddy (*J. Biol. Chem.*, 1920, XXXIV; see also *Am. J. Dis. Children*, 1917, 14, 189), has reported cases of marasmic infants which showed an increase in the utilization of food, when given vitamine extracts prepared from beans.

[4] In another connection the writer has found that mice and rats react differently with certain diets. The question is still open whether or not this difference is qualitative or quantitative.

from different sources. In this connection, it is believed that the procedure to be described later by the writer gives more direct evidence and hence is more free from criticism.

In order to eliminate the question of variability of individual animals from the point of view of efficiency as energy transformers, it appeared to the writer that a better procedure than that used by Hopkins would be to feed a number of animals a basal diet plus such an amount of vitamine-containing material as to keep the animal in weight equilibrium over a period of time. The amount of vitamine would vary with the individual, and would need to be determined in each case. When the animals had been maintained in weight equilibrium over a number of days and the food consumption noted, they could then be fed the same daily ration of a diet containing the same number of calories and having the same composition with the exception of a larger vitamine content and the weight noted. As the greater vitamine content would stimulate the appetite (directly or indirectly), there would be no difficulty about the animals eating the same amount of food as they had eaten in the first stage of the experiment and they would receive the same number of calories and a food with the same gross composition within very narrow limits.

Due to lack of time available for this work, it was impossible, in the preliminary experiment to be described, to bring each animal as near weight equilibrium as was wished, and as it is felt very certain can be done. The average, however, for twelve animals is close and if the results are considered from the statistical point of view, they give further very convincing evidence that the vitamines increases the efficiency of the body in the utilization of the food. The error due to temperature variation it is believed is not large, but this, of course, should have been eliminated. There appears no very easy way of eliminating the error due to greater activity of the animals in the second stage of the experiment. This error may be considerable for there is no question that the animals were markedly more active when given the limited diet with greater vitamine content. The direction of this error makes the results all the more convincing.

EXPERIMENTAL

Preparation of Diets.—The diets were prepared at the beginning of the experiment from the same stock of material and

placed in stoppered containers in the ice box. The composition follows:

	Diet 401	Diet 403	Diet 405
Casein	17.5%	16.5%	15.5%
Starch	49.5%	48.5%	47.5%
Yeast	1.0%	3.0%	5.0%
Lard	18.0%	18.0%	18.0%
Butter	9.0%	9.0%	9.0%
Salts	5.0%	5.0%	5.0%

The yeast contained approximately 0.46 per cent. fat, 46.5 per cent. protein and 38.0 per cent. carbohydrate (32.26 "carbohydrate" plus 5.8 crude fiber). Omitting the negligible quantity of salts the gross composition of the diets was as follows:

	Diet 401	Diet 403	Diet 405
Protein	17.96%	17.88%	17.8 %
Carbohydrate	49.88[5]	49.64[6]	49.4[7]
Fat	27.05%	27.15%	27.25%
Salts	5. %	5. %	5. %

[5] Includes 0.06 per cent. crude fiber.

[6] Includes 0.12 per cent. crude fiber.

[7] Includes 0.3 per cent. crude fiber.

The diets richer in vitamine had slightly lower calorific value.

The *casein* was prepared from the 40-mesh commercial product by shaking up for several hours successively with two portions of 50 per cent. alcohol and one of 95 per cent. alcohol and drying in warm air.

The *starch* was prepared from commercial cooking starch (corn) by the same procedure as for casein.

The *butter* used was the clear fat obtained by decanting the melted butter through a dry filter.

The salt mixture was that described by Osborne and Mendel.[8]

The mice were kept in wire mesh cages under which were pieces of glazed paper to catch feces and wasted food. The food was placed in small salve jars having aluminum covers in which ½ inch holes had been stamped. The jars were placed in 5-inch glass crystallizing dishes during the second stage of the experiment so that any waste food could later be found by the mice and eaten. Almost never in the second stage of the experiment was any food found under the cage. In all cases

[8] *J. Biol. Chem.*, 1919, 37, 572.

the waste was easily separated from the feces and account taken of the amount.

Sixteen mice were fed ten days on a complete diet and then nine days on a diet free of vitamines and then transferred to diet 401 (unlimited amount). After one day on the later intake records were begun.[9] Of the 16 mice started two grew nearly normally on 1 per cent. yeast,[10] one became sick and died, another declined very rapidly. These four were not considered in the experiment. The records of the remaining twelve are shown in the table. They were of varying size and represented somewhat different ages so that the results can hardly be accounted for by the action of the normal intermittent growth impulse.

PERIOD 1. DIET 401 (1 PER CENT. YEAST), UNLIMITED AMOUNT

Animal	Weights			Total Intake	Total Days	Average Daily Intake
	Beginning.	End	Change			
1.....	15.7 gr.	16.62 gr.	+0.92	27.15 gr.	13	2.09 gr.
2.....	11.2	10.49	−0.71	13.61	10	1.36
3.....	14.6	15.31	+0.71	24.75	13	1.9
4.....	11.1	10.45	−0.65	16.53	13	1.27
7.....	12.4	13.0	+0.60	19.83	13	1.52
8.....	14.0	14.97	+0.97	21.19	13	1.63
9.....	15.4	13.68	−1.72	19.39	13	1.49
10.....	14.8	13.71	−1.09	18.25	13	1.4
12.....	14.8	14.69	−0.11	21.91	13	1.68
13.....	18.5	17.05	−1.45	27.85	13	2.14
14.....	10.2	9.12	−1.08	13.77	13	1.06
17.....	15.9	16.58	+0.68	19.97	10	1.997
	168.6	165.67	−2.93	244.20	150	

For 150 mouse days 12 mice ate an average of 1.628 grs. per mouse per day and lost 2.93 grs. or 1.74 per cent. For maintenance, then, they needed slightly more than 1.628 grs. average per day.

Figured from Period No. 1 the mice in the 187 mouse days should have required a little more than 187×1.628 grs. and gained 14.36 grs. or 8.68 per cent.

[9] It would be well to increase this period somewhat.

[10] In general mice require 5 per cent. of yeast in the diet for normal growth; some make substantial gains on 3 per cent. and a very small per cent. require less. The individual variation for rats is, in our experience, considerable though not as great as for mice. The average requirement, too, for mice is markedly more than for rats, the ratio being roughly five for mice to three for rats expressed in per cent. in the diet.

PERIOD No. 2. DIET 403 AND (OR) 405 (LIMITED)

Animals	Beginnings	End	Change	Total Intake	Total Days	Fed Daily, Grs.
1.....	16.62 gr.	18.74 gr.	+ 2.12	32.64 gr.	16	2.04
2.....	10.49	11.8	+ 1.31	10.95	8	1.4
3.....	15.31	17.03	+ 1.72	30.40	16	1.9
4.....	10.45	11.83	+ 1.38	18.72	16	1.17
7.....	13.00	14.51	+ 1.51	24.00	16	1.5
8.....	14.97	14.05	− 0.92	24.00	16	1.5
9.....	13.68	14.45	+ 0.77	24.00	16	1.5
10.....	13.71	15.25	+ 1.54	24.00	16	1.5
12.....	14.69	14.65	− 0.04	25.6	16	1.6
13.....	17.05	18.84	+ 1.79	35.36	16	2.21
14.....	9.12	9.69	+ 0.57	17.00	16	1.06
17.....	16.58	19.21	+ 2.63	38.00	19	2.00
	165.67	180.05	+14.38	304.67	187	

In order to make more easily comparable the weight changes with food intake in the two periods, table No. 3 is presented.

Animal	Period No. 1			Period No 2		
	Weight Change	Average Daily Food Consumption	Days	Weight Change	Average Daily Food Consumption	Days
1.....	+0.92 gr.	2.09 gr.	13	+2.12 gr.	2.04 gr.	16
2.....	−0.71	1.36	10	+1.31	1.4	8
3.....	+0.71	1.9	13	+1.72	1.9	16
4.....	−0.65	1.27	13	+1.38	1.17	16
7.....	+0.60	1.52	13	+1.51	1.5	16
8.....	+0.97	1.63	13	−0.94	1.5	16
9.....	−1.72	1.49	13	+0.77	1.5	16
10.....	−1.09	1.4	13	+1.54	1.5	16
12.....	−0.11	1.68	13	−0.04	1.6	16
13.....	−1.45	2.14	13	+1.79	2.21	16
14.....	−1.08	1.06	13	+0.57	1.06	16
17.....	+0.68	1.997	10	+2.63	2.00	19

The energy content of the excreta during the two periods was not determined, but Hopkins[11] and Drummond[12] have shown that the energy content of the excreta of rats when fed adequate foods is substantially the same as when fed vitamine-free foods.

The writer regrets the inability to repeat the work with a larger number of animals. It would be desirable to use a vitamine extract, control the temperature of the animal room, get the animals more closely in weight equilibrium by using a vitamine percentage determined by the individual mouse, and to

[11] *J. Physiol.*, 1912, 44, 440.

[12] *Biochem. J.*, 12, 25.

have periods 1 and 2 run over the same number of days. Among the animals there should be included several adults which had been brought to condition of underweight in the preliminary period with vitamine-free foods. This would answer any possible question concerning the effect of the intermittent growth impulse. It is hoped that others may be able to carry on this work.

These experiments were performed under a grant from the Committee of the Permanent Charity Fund, Incorporated.

R. R. RENSHAW

HARVARD MEDICAL SCHOOL, BOSTON

INTERFERENCE IN PRIMULA SINENSIS

OUTSIDE of *Drosophila*, the only data bearing on the question of interference of crossing over are those which I reported in a paper on linkage in *Primula sinensis*.[1] The phenomenon of interference—our knowledge of which in *Drosophila* dates from crosses made by Sturtevant and analyses made by Muller in 1912—consists of the fact that the occurrence of a crossing over in one region of a chromosome reduces the chances for the occurrence, in that cell, of another crossing over in a different region of the same chromosome; thus there is a smaller number of double crossovers than would otherwise be expected. The amount of interference is expressed by Muller's index called "coincidence," which is the ratio of the proportion of double crossovers actually observed in the experiment to the proportion of double crossovers which would have been expected to occur on the assumption that crossings-over in the two regions were independent of each other; the latter, or "expected" proportion of double crossovers is obtained by simply multiplying together the proportion of crossovers in one region by the proportion of crossovers in the other region. As I stated in my paper on *Primula*, a calculation based upon my total results could not be sufficiently reliable to decide the question of whether or not interference existed in *Primula*. This was on account of an uncertainty in the classification; I now find, however, that a calculation based upon a selected group of the plants, in which the uncertainty does not exist, is sufficient to decide the question in the affirmative—contrary to my earlier conclusion.

Three pairs of genes were involved in the *Primula* crosses—

[1] Altenburg, E., 1916, Linkage in *Primula sinensis*. *Genetics*, 1: 354–366.

those for long style (l), red flower (r), and red stigma (s), allelomorphic respectively to short style (L), magenta flower (R), and green stigma (S). The order of the loci, as based upon 3684 individuals, was l r s; the per cent. of crossovers in the first region (between l and r) was 11.62, and that in the second region (between r and s) was 34.02. These relations, shown in a map, are as follows: $\frac{l}{0}\ \frac{r}{11.62}\ \frac{s}{45.64}$. The per cent. of double crossovers observed in the experiment was 2.52. According to the formula given above, the number of double crossovers to be expected if crossings over were independent would be 11.62 per cent. $\times$ 34.02 per cent., or 4.0 per cent., which, as I noted in the account of the case, exceeds the observed proportion of 2.52 per cent. This difference, then, between the observed and "expected" values in *Primula* would indicate that interference existed here, but, as I further stated, the difference was not significant because of the uncertainty which had attended the classification of flower color in the plants with the gene for green stigma. This gene caused the flower color to be lighter and obliterated somewhat the distinction between red and magenta.

In the plants with red stigma, however, the flower color was dark enough to render entirely certain the classification in regard to red and magenta; these plants, considered alone, would therefore furnish reliable data for determining the interference. I stated that, when these reliable plants alone were taken into account, no evidence of interference was to be found; but this conclusion was due to a numerical error in the calculation of the "expected" number of double crossovers, for I now find, in going over the figures, that the "expected" number is considerably higher than the number observed. Among the 1876 plants with red stigmas, there were 210 crossovers, or 11.2 per cent., in the first region, and 688, or 36.7 per cent., in the second region. The "expected" number of double crossovers is therefore 11.2 per cent. $\times$ 36.6 per cent., or 4.1 per cent. There were 54 double crossovers observed, or 2.9 per cent., giving a coincidence ratio of 2.9 : 4.1, or .7, instead of 1.00, which would be the ratio in the absence of interference. The difference between the "expected" and observed numbers is beyond the limits of random sampling, and it must therefore be concluded that interference exists in *Primula*. Although no reliance can be

placed upon the precise value of the coincidence, it may be noted that this amount, .7, is just what would be expected for a similar distance in the X chromosome of *Drosophila melanogaster* (*ampelophila*).

It should be noted here that Haldane, in referring to my results in a recent article, called attention to the fact that the *Primula* data (using the counts of all classes of plants) fit his formula for expressing the relations between linkage values in *Drosophila*. Inasmuch as any formula expressing the linkage relations in *Drosophila* is necessarily the mathematical resultant of the operation of interference (interference of a type which diminishes with increasing distance), Haldane's statement that the *Primula* data fit the same formula as *Drosophila* is equivalent to saying that interference exists here, as in *Drosophila*; it is in this sense a restatement of my observation that the number of double crossovers found in the total count of the plants is smaller than the number "expected" in a case of a random occurrence of crossing over. It must further be noted that Haldane's formula for expressing the linkage relations in *Primula* is open to the same objection of unreliability as noted above, since his calculation is based upon all classes of plants, instead of upon just those classes which I showed must be used in any reliable determination.

The finding of interference in another organism, so widely separated from *Drosophila* is of interest because of the bearing of interference on the general theory of linkage. Interference is not accounted for on Trow's form of the reduplication theory, although it was the earlier experiments upon *Primula* itself which largely supplied the data upon which this theory was founded.

EDGAR ALTENBURG

THE RICE INSTITUTE,
HOUSTON, TEXAS.

ON INTERSEXES IN FIDDLER CRABS.

NOT long ago a few specimens of small female fiddler crabs, *Uca pugnax*, were submitted to me by Professor T. H. Morgan for determination. They appeared to be normal, immature individuals and I so stated in my reply. In order to demonstrate the correctness of this view, a loan was made to Professor Morgan from the National Museum collection of a series of imma-

ture female fiddlers showing different widths of abdomen normal to the growing female in passing from the juvenile or immature, to the sexually mature, condition. However, this exhibit apparently had the opposite effect from that intended, as Professor Morgan pronounced them all "intersexes" and thereby seemingly robbed the female fiddler of its period of adolescence.

In his paper "Variations in the Secondary Sexual Characters of the Fiddler Crab"[1] we find Professor Morgan's exposition of the subject. It is not easy to follow the author owing (1) to erroneous or incomplete references to figures and (2) to absence of measurements; for example, "Fig. 4*B*″," cited on p. 225, line 12, does not exist, "Fig. 4*B*′," p. 225, line 3, is cited as a female abdomen when it is really a male, and one can not tell if the two unequal chelæ of Fig. 4*A* belong to one individual, which is probable, and if the two unequal chelæ of Fig. 4*B* belong to one individual, which is probable but not possible, as the text says that they are "of the same size." There are no measurements nor indication of enlargement of figures.

The case under discussion belongs in Professor Morgan's second category of intersexes. He tells us that the specimens are always small, that they are female in character except for the abdomen being narrower than in the mature female, and the abdominal appendages being different from those of the mature female but not at all malelike. Why, one naturally asks, are they not juvenile? His reply is, "because normal individuals of the same size have the abdomen full width." This argument unsupported is fallacious.

Many species of crabs are known to attain sexual maturity at a much smaller size than their maximum and to exhibit considerable range in the size at which they attain that maturity. As an example, two jars full of the common shore-crab of the Pacific coast, *Hemigrapsus nudus*, show egg-bearing crabs ranging in width of dorsum from 10.4 mm. to 32 mm., and among the immature females with narrow abdomens, six individuals which range from 12.5 to 15.7 mm. in width.

Professor Morgan goes on to say that some of the smallest "intersexes" have the narrowest abdomen, that there is no obvious relation between the size of the crab and the relative width of the abdomen, but that there is some correlation between the character of the abdominal appendages and the width of the

[1] AMER. NAT., Vol. LIV, No. 632, May-June, 1920, pp. 220–246.

abdomen. All these point to normal development as the rational explanation.

He figures, p. 226, the abdomens of five female "intersexes," including, I think, but am not sure, two abdomens of successive, or near successive molts in the aquarium. As no two of these abdomens are of the same width, the illustrations would indicate a change in size of body, that is, growth and surely age, with the molt or molts. But Professor Morgan says, p. 225, lines 13–14, "that the condition of the abdomen and claws had not changed."

The fact of the matter is, that neither Professor Morgan nor any one else, so far as I know, is aware of the exact growth changes of our fiddler crabs beyond the first few crab stages. Hyman, in "The Development of *Gelasimus* after Hatching,"[2] carries his painstaking researches only as far as a 4 mm. wide crab stage.

We can at present reason only by analogy from the study of work done on other species of crabs, of which there is altogether too little compared to the facilities offered by the laboratories of our coasts; and such analogy seems to indicate that the crabs upon which Professor Morgan bases his arguments are normal females which had not, in their particular cases, attained sexual maturity. Churchill's "Life History of the Blue Crab"[3] may be cited, and also Pearson's "Cancer. (The Edible Crab.)"[4] Both of these give tables which demonstrate the great variability in the ratio of increase at each act of ecdysis.

It is important, as I have stated elsewhere, that the development of each of our common crabs be carried through from the egg to maturity, that accurate records be made, and properly labeled material upon which such studies are based be deposited in an enduring collection accessible to all who may be interested.

MARY J. RATHBUN

VARIATION IN JUVENILE FIDDLER CRABS

IT is too bad that Miss Rathbun's kindness in sending me specimens from the National Museum "had the opposite effect from that intended." While regretting this, I can only call attention to the fact, stated in my paper, that out of more than

[2] *Jour. Morphol.*, Vol. 33, No. 2, March, 1920.

[3] *Bull. Bur. Fisheries*, XXXVI, November 11, 1919.

[4] *Proc. and Trans. Liverpool Biol. Soc.*, Vol. XXII, 1908.

three thousand individuals that were collected only a few showed the narrow abdomen concerning which Miss Rathbun raises an interesting question. These rare individuals, I ventured to suggest, with some hesitation and with considerable reservations, might be called intersexes, because the variation in question was in the direction of a character peculiar to the opposite sex. Miss Rathbun states that my argument that "they are not juvenile . . . unsupported, is fallacious." Again I can only repeat what was said in my paper, that I examined a very large number of individuals, many of which were of about the same size as the variations in question, some of which were even smaller, and others somewhat larger, and in none of the young females (except in those recorded as exceptions) did I find the abdomen narrow.

May I also recall that I specifically referred to the case of the blue crab in which the abdomen of the juvenile female is narrow, so this condition was known to me, both from the literature and from personal examination, although the reader might gain the opposite opinion from Miss Rathbun's comments.

It is rather strange also that Miss Rathbun neglects to point out that these small crabs with narrow abdomen were stated in my paper to show either a change towards maleness or possibly a retention of the juvenile condition. It is quite possible, of course, that the narrowness of the abdomen of the exceptional individuals might be interpreted as a variation in the direction of the juvenile stage found in other species; but it is certainly not a common stage through which crabs of this size pass. Whether the juvenile interpretation has any advantage over the alternative one that I provisionally suggested can only be settled when we have found out to what this exceptional condition in the fiddler crab is due. My paper was written more with the intention of calling attention to a new and very interesting set of variations in these crabs (as the title indicates) than with the intention of trying to determine what the definition of intersexes shall include; for, as I said, "It seems to me not worth while at present to attempt to classify such material until we have learned more about it."

T. H. MORGAN

[1] Fig. 4*B'*, page 225, line 3, should be Fig. 5*B'*. Fig. 4*B''*, page 225, line 13, should be Fig. 5*C''*. Fig. *A, B, C, D*, page 225, line next to bottom, should be Fig. 5 *A, B, C, D*.

THE TURKEY AS A SUBJECT FOR EXPERIMENT

EXPERIMENTS with our native vertebrates offer many difficulties not encountered when dealing with domestic animals. In the field of genetics especially, while domestic animals continue to furnish enticing problems, it is not strange, therefore, that they practically monopolize the attention of students. No one can foresee how far work of this kind will proceed but it seems probable that some important phases of the subject of variation never can be elucidated by the study of domestic animals alone. Hence it would be very desirable to work with wild forms wherever this is practicable. This would be especially interesting for study of the significance of the intergrading subspecies or "geographic race" which is found so widely in nature but which appears to have no recognizable counterpart in the ordinary variations of domestic animals.

The so-called subspecies perhaps needs no introduction even to biologists who do not have first-hand acquaintance with it, but the extent to which it features in the fauna of the world seems scarcely realized even among those who are quite familiar with it. Within the memory of the present generation, the ultimate division of classification was the species and attempts to divide this into races or varieties were often looked at askance as probably indicating an over-weening desire to multiply names and magnify differences of no phylogenetic significance. In wrestling with the question "What is a species?" many were led to eschew classification entirely and contented themselves with the knowledge that no two individuals were alike and the belief that efforts to associate them were futile. Meanwhile, in spite of headshaking in various quarters, "hair-splitting" has continued until at present nothing is clearer than that the subspecies is a reality constituting a widespread and obvious evidence of active contemporary change in organisms, not only in single individuals but in groups of individuals.

In ornithology and in mammalogy, at least, the old-fashioned species in the vast majority of cases is found to be a composite or a mosaic definitely divisible into units connected by graded series and having a plain relation to geographic distribution. A species of continental distribution in North America, for example, may have one subspecies in the east, one in the north and several in the south and west each occupying a limitable area and each characterized throughout its range by certain features

not possessed by the others. Along the geographic borders of each subspecies will be found specimens showing varying degrees of intergradation so that each form merges with an adjoining one, or, in some cases, one in central position may merge into several others in different directions. If, for any reason, the "areas of intergradation" were rendered uninhabitable, the various subspecies would stand as distinct well-characterized forms presumably until they themselves began to differentiate and separate into parts. Sometimes the difference between recognizable subspecies is slight, or sometimes it is very marked, but when gradations through several forms are followed, characters are almost always found to change to a degree far beyond any probability of an ontogenetic explanation. It may be emphasized that subspecies of this sort are not the exception, but the rule. It might almost be said that the existence of diverse inosculating units correlated with geography is characteristic of terrestrial vertebrates. Continued study with improved facilities and increasingly comprehensive collections from all parts of the world constantly reduces the number of forms which are not known to break up into subspecies. To a very great extent, the presence of an undivided 'full species" in our checklists signifies either that it is a senescent type of limited distribution or that, for lack of material or opportunity, it has not been studied intensively. The intergrading subspecies has not been recognized so widely among invertebrates nor in plants, but neither entomologists nor botanists have collected and studied their material from the geographic standpoint to such an extent as the ornithologists and mammalogists, so it cannot be said that the process of change illustrated by the subspecies is not even more widespread than appears from the study of birds and mammals.

The process of formation of these subspecies, therefore, is going on before our very eyes in wholesale fashion and it is difficult to believe that it is, as someone has said, merely a "shuffling of the cards" which in the long run means nothing to evolutionary progress. Rather does it seem that it must have a physiological basis, a relation to germinal change, and a large potentiality for affecting the general course of evolution. Despite its evident importance, the intergrading subspecies is receiving but scant attention from experimental zoologists. With the conspicious exception of the very significant work being done with

white-footed mice by Dr. F. B. Sumner of the Scripps Institution,[1] there seems to be little or no work under way which can be correlated logically with the results of speciation and subspeciation as the field naturalist and taxonomist find them in nature. Doubtless one of the principal reasons for this is the difficulty of finding convenient subjects and suitable conditions for such work. Perhaps another is the independence of workers in the respective fields of taxonomy and experimental zoology.

As promising subjects for experiment, it seems worth while to call attention to the American wild and domestic turkeys. The common turkey has an exceedingly desirable distinction from other domestic animals in that there is no important question as to its history and lineage. Moreover, the wild stock from which it was derived represents one of several intergrading subspecies the natural characters and relationships of which can be determined with a great degree of accuracy. Hence our Thanksgiving bird, as a subject for experimental breeding, might furnish a combination with which naturally and artificially induced characters could be studied comparatively. As at present recognized and understood, the native American turkey is divisible into six races or subspecies, as follows: One from the southeastern United States (*Meleagris gallopavo sylvestris*); one from southern Florida (*M. g. osceola*); one from central Texas and northeastern Mexico (*M. g. intermedia*); one from Arizona, New Mexico and Chihuahua (*M. g. merriami*); one from the Sierra Madre of Jalisco and west central Mexico (*M. g. mexicana*); and one from the eastern cordillera of Vera Cruz, Mexico (*M. g. gallopavo*). The range of the turkey group is thus from the southeastern Atlantic seaboard westward to the Rocky Mountains and thence south to Vera Cruz. Complete intergradation between the various subspecies may not be demonstrable with absolute nicety in all cases because the birds were exterminated in certain parts of the range before any specimens were preserved. That intergradation between all the races was as uninterrupted as it can be shown to be between some of them, however, is beyond reasonable doubt. The extremes of differentiation, as usual in such cases, are represented approximately by the geographical extremes. The characters distinguishing the wild turkey of the eastern United States from that of south-

[1] See especially AM. NATURALIST, XLIX, pp. 688–701; *ibid.*, LII, pp. 177–454, 1918.

ern Mexico, therefore, are clear cut and readily recognizable without the application of any greatly refined methods. The obvious distinction is found in the feathers of the tail and upper tail coverts which in the United States bird are broadly tipped with rich chestnut whereas in the Mexican subspecies these parts are white or nearly white. Such characters, in animals of unknown history, might easily be looked upon as produced by mutation; but with complete gradation from one to the other known to exist in nature, it is hard, at least for some of us, to believe that the difference was not accomplished by gradual rather than sudden change. If it could be shown that characters of this kind behave as hereditary units without any such blending as requires "dialectic gymnastics" to explain, it would be a long step forward in the correlation of natural and man-made experiments. Such characters are in fact heritable, as has been shown by Sumner in his breeding and transference experiments with *Peromyscus*. This is illustrated also by an undirected experiment to call attention to which is one of the objects of this communication, namely, the test of subspecific characters which has been carried out in the domestication of the turkey.

As is widely known to sportsmen, breeders, and many others, our domestic, so-called bronze, turkey is readily distinguished from the wild bird of the eastern United States by the coloration of the upper tail coverts and tail. The reason for this, which is not so generally known, is not that the domestic bird has changed in these respects under man's influence, but because it is the direct descendant of the Mexican wild race which differs from the northeastern race by these selfsame characters. Carried from Mexico to Europe in the early sixteenth century and thence brought to the United States, it has continued for more than three hundred generations in a new environment maintaining its old established subspecific characters. To-day it may differ from the Mexican wild race in some details, but its general coloration is the same and especially does it retain its taxonomically diagnostic features. These, therefore, are heritable and doubtless related to germinal conditions which became fixed in the wild bird. Since the characters themselves are of the kind that appear to be produced by insensible gradations and of the kind that frequently bear an obvious relation to environment. an easy deduction would be that the germ plasm also has

changed gradually and, at least as a working hypothesis, one might suppose that the germ plasm had been affected directly or indirectly by the environment. Such an explanation is, of course, far too simple and old-fashioned for present-day students of evolution. It does not explain a multitude of undeniably important and fascinating results of experimental work. But neither do theories of mutation and the maze of modern genetics explain the intergrading subspecies and perhaps there is room for at least a little experimental work which does not deny such an hypothesis at the outset.

In the case of the turkey, while the relatively inconspicuous subspecific character has proved itself stable, violent saltatory changes have been established easily. These are of the sort common among close bred domesticated animals but so rare among wild vertebrates that no one has yet found a case in which they can be shown to have been perpetuated by natural process. Thus we now have self-colored breeds of turkeys respectively black, white, buff, and blue gray as well as the breed called Narragansett in which the feathers are tipped with steel gray. Hence it seems that the turkey may offer an opportunity for comparative study of the hereditary behavior of characters which have developed naturally by what seems to be continuous variation and those which have appeared discontinuously and been perpetuated artificially. Sumner (l. c., 1918) has found with *Peromyscus* that hybridization of different subspecies produces in the F_1 and F_2 generations a blending of the subspecific characters comparable to the gradations found in nature, whereas mutant characters (partial albinism, etc.) act as simple Mendelian units. In other words, natural subspecific characters act in hybridization experiments as they would be expected to do on the assumption that they were produced by continuous variation. Whether or not the same results would follow with the turkey and other forms would seem to be well worth determining. In a general way, it is known to breeders that hybrids between wild and domestic turkeys are of intermediate type, but so far as I know careful well-controlled work has not been done.

WILFRED H. OSGOOD

FIELD MUSEUM OF NATURAL HISTORY

THE "ONE-LETTER" RULE FOR GENERIC NAMES IN ZOOLOGY

RULES of nomenclature as they affect scientific names in zoology would no doubt serve their real purpose best and give more general satisfaction if used, not in an absolute sense, but with discretion. But there are those to whom a rule is a rule to be rigidly applied, and the results are such that the question is raised whether if we must abide by rules, we cannot have better ones. A nomenclatorial rule used by the American Ornithologists' Union has raised this question in the mind of the writer, and attention is here called to it, not in a controversial way, but merely to insure that the other side of the case is presented. If such questions are ever taken up again by an International Commission it is desirable that data and opinions on the vexed points be available for consideration. In the recently published[1] "Seventeenth Supplement to the American Ornithologists' Union Check List of North American Birds," prepared by the Committee on Nomenclature, we find the following statements relating to certain generic names:

Oxyura Bonaparte 1828 is considered preoccupied by *Oxyurus* Swainson 1827 (p. 446).

Nyctala Brehm, from whatever date taken, is preoccupied by *Nyctalus* Bowditch 1825, and *Ægolius* Kaup, 1829, is preoccupied by *Ægolia* Billberg, 1820 (p. 447).

Bucephala Baird 1858 is preoccupied by *Bucephalus* Baer 1827 (p. 446).

Dendrocopos Koch, July, 1876, is preoccupied by *Dendrocopus* Vieillot, April, 1876 (p. 448).

But

Heteroscelus Baird 1858 is not invalidated by *Heteroscelis* Latreille 1825 (p. 443).

Tyto Billberg (1828) is not preoccupied by *Tyta* Billberg (1820) (p. 447).

and

Moris Leach . . . adopted because considered neither a *nomen nudum* nor preoccupied by *Morum* Bolten, although *Morus* Vieillot . . . having a termination differing merely in grammatical gender from *Morum* Bolten is thereby invalidated (p. 441).

[1] *The Auk*, Vol. 37, No. 3, July, 1920, pp. 439–449.

Even the experienced taxonomist might be greatly puzzled by this collection of apparently inconsistent assertions, did he not turn to the Code of Nomenclature of the American Ornithologists' Union (1908 Edition) and find the following explanatory remark under Canon XXX:

> Generic and specific names . . . are to be considered identical . . . whether the ending is masculine, feminine or neuter or in Greek or Latin form.

In the principal codes of zoological nomenclature the practise called for by this rule is sanctioned only by that of the American Ornithologists' Union. The parent (we may say) of the A. O. U. Code, namely the Stricklandian Code, in so far as it touches on the point, would seem to accept very similar generic names, even those differing by only one letter. Section 10[2] says

> A name should be changed which has before been proposed for some other genus in zoology or botany.

This section is further elaborated as follows:

> By Rule 10 it was laid down, that when a name is introduced which is identical with one previously used, the latter one should be changed. Some authors have extended the same principle to cases where the later name, when correctly written, only approaches in form, without wholly coinciding with the earlier. We do not, however, think it advisable to make this law imperative, first, because of the vast extent of our nomenclature, which renders it highly difficult to find a name which shall not bear more or less resemblance in sound to some other;[3] and, secondly, because of the impossibility of fixing a limit to the degree of approximation beyond which such a law should cease to operate. We content ourselves, therefore, with putting forth this proposition merely as a recommendation to naturalists, in selecting generic names, to avoid such as too closely approximate words already adopted (p. 118).

These provisions were adopted (with a reservation as to botanical names) by the British Association for the Advancement of Science in 1865 as part of a code which more than any other guided the course of subsequent nomenclature practice.

[2] Rep. British A. A. S., 1842 (1843), p. 113.

[3] If this was true in 1842, how much more difficult the situation must be now after 80 additional years of taxonomic activity.

In Dall's "Discussion of the Subject of Nomenclature" of 1877 which was based on a circular responded to by 45 American naturalists in addition to previous codes and other publications on the subject, the point under consideration receives the following attention in Section 65, Paragraph 10,

> When a name is identical, when properly spelled according to a derivation given by its author, with a prior valid name in the same kingdom it must be rejected.[4]

In other words, if names are not *identical* they stand.

The Entomological Code (1912, Paragraph 82) has this to say on the subject:

> A generic or subgeneric name is a homonym and subject to replacement when it is spelled exactly like a previous valid generic or subgeneric name, letter for letter. However, I and J, and Eu and Ev at the beginning of a name are considered the same, and other words that are equivalent in established Latin usage.

In extracts from a code of Nomenclature in Ichthyology (Jordan, Evermann and Gilbert) published in the Condor in 1905, Canon XVII (Second paragraph), is quoted as follows:

> As a name is a word without necessary meaning, and as the names are identified by their orthography, a generic name (typographical errors corrected) is distinct from all others not spelled in exactly the same way. Questions of etymology are not pertinent in case of adoption or rejection of names deemed preoccupied. (Note.) This canon prohibits change of names because prior names of similar sound or etymology exist. It permits the use of generic names of like origin but of different genders or termination to remain tenable.

The International Code which, so far as it goes, is adhered to by a majority of zoologists, alludes to this subject in a recommendation under article 36. The language follows:

> It is well to avoid the introduction of new generic names which differ from generic names already in use only in termination or in a slight variation in spelling which might lead to confusion. But when once introduced, such names are not to be rejected on this account.

[4] Nomenclature in Zoology and Botany, Salem, Mass., December, 1877, p. 49.

Opinions 25 and 34 of the International Commission support the wording of the foregoing recommendation which is referred to in the opinions as an effective part of the code.

Thus zoological codes in general support the so-called "one-letter rule." The point in this connection that appeals to the present writer with special force is that there would seem to be no good defense for the practise of rejecting names differing in terminations expressing gender and at the same time accepting other names differing by no greater margin (often by only one letter).

Thus under A. O. U. practise *Otostomus*, *Otostoma* and *Oto stomum* are treated as identical, while *Odostoma* and *Otostoma*, *Icteria* and *Icterias*, *Pica* and *Picus* are considered distinct. The fact that the latter words had different terminations, or different meanings in classical usage has nothing to do with the case. Nomenclature is not the Latin language; it is a mass of invented, adopted, derived and compounded words, some of which are in Latin form, others not, but all of which, nevertheless, have equal standing in the scientific world. Principle V of the A. O. U. Code, itself, asserts that

> A name is only a name, having no meaning until invested with one by being used as the handle of a fact; and the meaning of a name so used in zoological nomenclature, does not depend upon its signification in any other connection.

Literally construed this principle is fully in accord with the definition of scientific names as arbitrary combinations of letters, and it would seem unnecessary even to state with respect to arbitrary combinations, that we can only regard each different one (even if by only one letter) as a distinct name. It would seem clear, therefore, that in scientific nomenclature names are merely labels for conceptions; that their use demands precision, and with precision all names appreciably different can be used without confusion.

Small (even one-letter) differences in scientific names are by no means confined to terminations; they occur in all points in words. Consider: *Neothripa, Neothrips; Felicea, Felicia, Donatia, Donacia; Isotoma, Isosoma; Leptopora, Leptoprora; Mercera, Merciera; Teliocrinus, Teleiocrinus; Sciurus, Seiurus; Sus, Mus.* Consider also such a series of names as *Monocerus, Monocereus,*

Monocercus, Monocercis. These names all stand under the A. O. U. Code, as do also words like the following: *Rolanda, Rolandra; Oga, Ogoas; Orophia, Orophila; Menida, Menidia; Lyria, Lyrcia; Passerina, Passerita;* all of which differ only in the last few letters as do those with terminations denoting gender, and are equally liable to confusion by typographical errors.

What justification is there for accepting names so nearly alike as many of these but considering as homonyms such terms as *Nyctala* and *Nyctalus; Nettion* and *Nettium?* The aim of codes of nomenclature is to conserve names, not to make opportunities for the creation of new ones. But the A. O. U. custom of considering homonyms, names differing in terminations indicating gender is a breeder of new names. This is clearly shown by two notes[5] published in a recent number of *The Auk,* in which it is asserted that *Phæochroa* Gould 1861 is preoccupied by *Phæochrous* Laporte, 1840, and *Elminia* Bonaparte 1854 by *Elminius* King, 1831, and a new name is proposed in each case. The same criticism applies to certain other suggestions in connection with Canon XXX of the A. O. U. Code, namely those that would homonymize such words as *Athene* and *Athena; Contopus* and *Contipus.* Those who look with favor on homonymizing words whether they differ only by endings denoting gender, whether the root is taken from the Attic or other dialect, whether the connecting vowel of compound words be a, i, or o, or for other philological reasons should remember that there is no more reason for stopping at one point than another in the path of purism. Always there will be more and more advanced purists, who would sink generic names differing far more widely.[6] For instance, consider the following pairs of names for which it has actually been proposed that the second name in each couplet be regarded as homonym: *Callitriche, Calothrix; Myosuros, Myurus; Galarhoeus, Galactorheus; Korycarpus, Corythrocarpus; Ionactis, Iactis; Genyscoelus, Coelogenus.*

Philology is an interesting and important science, but what has classical purism to do with a hodge-podge of names such as zoological nomenclature now is, with names coined, with names classical, with those borrowed from nearly every language ancient and modern? What would be the fate of nomenclature if the purist were allowed to work his will with such names as: *Abudef-*

[5] *The Auk,* Vol. 37, No. 2, April, 1920, p. 295, and p. 302.

[6] A very few reject words of similar sound—phononyms.

duf, Avahi, Aye-aye, Bagre, Cachalot, Djabub, Grysbock, Jafar, Jukaruka, Kahavalu, Louti, Mabuya, Maki, Ompok, Potto, Sandat, Sheltopusik, Tlja, Susu, Wallago, Zingel and the like? Or with such personal and local derivatives as: *Amiskwia, Ernestokokenia, Ischikauia, Mitsukurina, Mordwilkoja, Schlaginhaufenia, Takakkawia, Wankowiczium, Wlassicsia and Zschokkeella?*

The writer does not defend the choice of such names, but once on record they are an integral part of nomenclature and an outburst of purism sufficient to do away with them will not occur. Whether we will or no, we are dealing with essentially arbitrary combinations of letters arbitrarily selected. The conglomeration of generic names in zoology, may be, nay is, subject to criticism, but it exists, is in use. It is part and parcel of the language of Science and classical purism can no more be applied to it than to any other modern language which is constantly growing, ever adding to itself terms from a multitude of sources.[7] A condition not a theory confronts us; practicability must reign and pedantry be forgotten.

Practically all rules relating to the validity and priority of generic names have some saving clause as "typographical errors corrected," or "except for obvious typographical errors." A common-sense application of such clauses would do away with the most vexatious cases of emendation, cases often cited to show the necessity of homonymizing similar generic names, namely those in which an author mis-spells names of his own establishing when using them subsequently to the original citation. In such cases why can we not take an author at his word; he intended to treat of the same group as before, and his emended name, whether intentional or not should be regarded as a synonym of the original. We do not recognize an author's efforts to change a published name, except to correct typographical errors. Why should we give any weight to emendations which themselves, in many if not most cases, are almost certainly typographical errors. The same rule should apply to names mis-spelled by others than the original author when it is clear they intended to refer to the same genus. The fact that the species included under such names are now considered to belong to different genera is of no consequence; these genera

[7] Thus we adopt into English but do not Anglify such words as hangar, machete, fez, mufti, a host of which could be cited.

should date from the time formally recognized and should bear the name then given. It is a travesty on priority to credit an author with conceptions he never entertained, and to use for them mis-spelled names for which he no doubt often had occasion to regret his inadvertence. In brief, regard all emendations as typographical errors unless there is definite evidence to the contrary. With the treatment suggested, such cases as *Pogonius, Pogonias, Pogonia* (a name spelled three ways in the same publication), and similar instances lose their troublesome aspect, and suggestions for homonymizing them, much of their force. The chief cause for anxiety in connection with the one-letter rule seems to be that numerous emendations may be revived, but it can confidently be asserted that, from a practical viewpoint, most emendations are clear synonyms from the beginning and their status would not be changed under the one-letter rule.

Moreover changes under this rule need be feared only in branches of zoology in which the practice advised by the A. O. U. Code has been followed, that is the study of birds and mammals. Certainly the one-letter rule has been used, since the adoption of the International Code, if not before, by most American students of animal parasites,[8] echinoderms, crustaceans, insects and fishes[9] and as shown in preceding paragraphs their practice[10] in this respect is overwhelmingly supported by the various zoological codes.

[8] See discussions by Ch. Wardell Stiles (*Zool. Jahrb.*, 15, 1902, pp. 172–175). "The difference of a single letter, entirely regardless of the etymology, excludes the possibility of the words being identical, hence excludes the possibility of their being homonyms" (pp. 172–173).

[9] See note in Jordan and Evermann, "The Fishes of North and Middle America," Vol. I, 1896, p. v, "We regard all generic names as different unless originally spelled alike."

[10] An attempt to develop what usage, in this respect, is followed in a larger number of zoological specialties, was made by mailing a brief questionnaire to 30 systematic zoologists. The questions asked were:

1. In your specialty have one-letter differences been regarded in recent years (at least since adoption of International Code) as sufficient to establish the distinctness and validity of generic names?

2. Or has the ruling of American Ornithologists' Union Code relating to homonymizing terms differing only in endings indicating gender, etc., been followed?

Only 17 replies were received, of which 2 were noncommital, 9 reported no established usage and those which indicated adherence to one or the other of the opposed practise numbered 3 in each case. The result of this mail test at least supports the writer's contention that the subject is one ripe for public discussion.

Since one-letter differences in generic names are sufficient in many cases as shown by citations in this article and the practise of nomenclators, why are they not in all? The one-letter rule is practicable, while one based on grounds of classical purism is not, and as the framers of the Ichthyological Code properly remark:

> If all names are regarded as different unless spelled alike, these matters offer no difficulty. Any other view gives no assurance of stability.

Finally, discarding names of independent origin and distinct application, that are not spelled identically, overthrows the law of priority and like all practises of that tendency (so long as the priority system is followed) is not for the lasting good of scientific nomenclature.

W. L. McATEE

U. S. BIOLOGICAL SURVEY